Fundamentals of Crystallography

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First: What is your background?

Think about where you fit in these lists:

Graduate student **Post Doc** **Beyond Post Doc**	**Math** **Physics** **Chemistry** **Biology** **Medicine**

Outline for the Lecture

- Remind you how much you already know -- lenses, crystals
- Show why crystals give diffraction spots.
- Develop the idea of "The Reciprocal Lattice"
- Give some idea how we might actually measure diffraction data
- Show how, given a crystal, we can calculate the diffraction pattern
- Conversely, show how to calculate the structure from the diffraction
- Describe the importance of symmetry to diffraction
- Outline the structure-solving methods -- heavy atoms and MADness

The idea here:

- Firstly, we’re going to try to understand how things work.
- Then we’ll try to use that understanding to figure out how to solve a few problems one might meet in doing crystallography.
- Perhaps then you’ll be able to solve ***many*** problems in crystallography.

The idea here:

- We’re not going to spend time working through how to solve problems in crystallography.
- Instead, we’re going to try to understand how things work.
- If we can understand, then we can figure out how to solve almost any problem.

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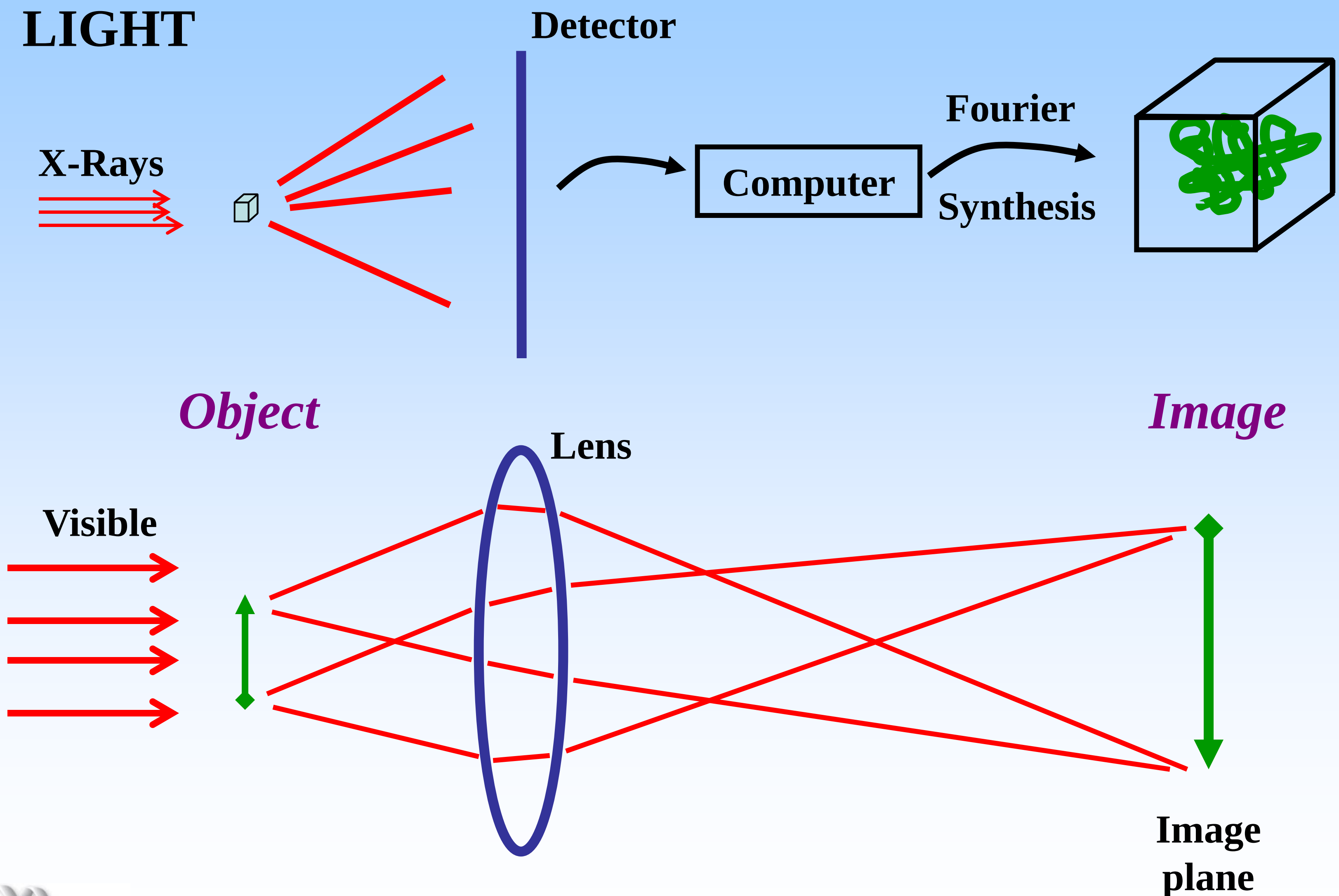
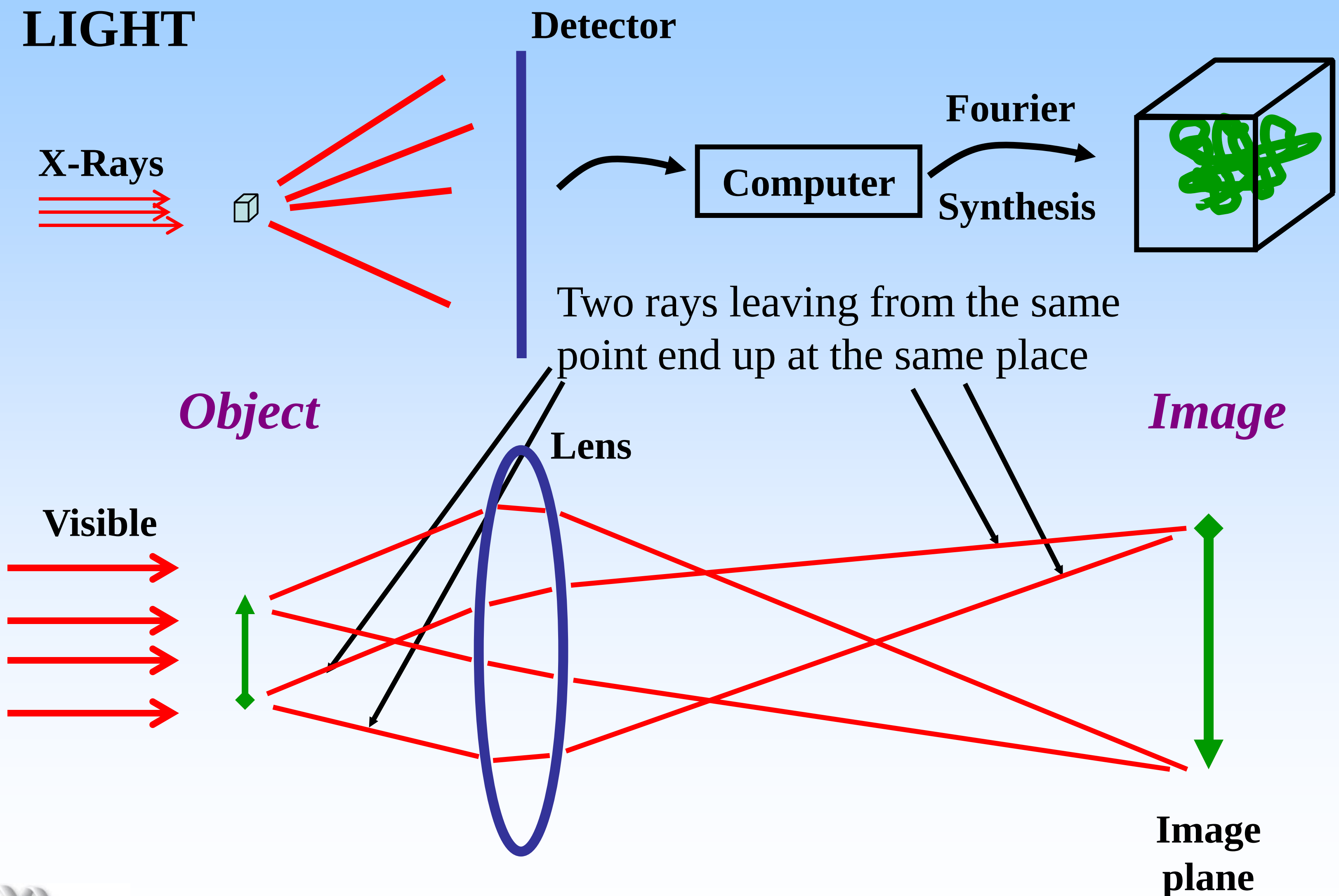
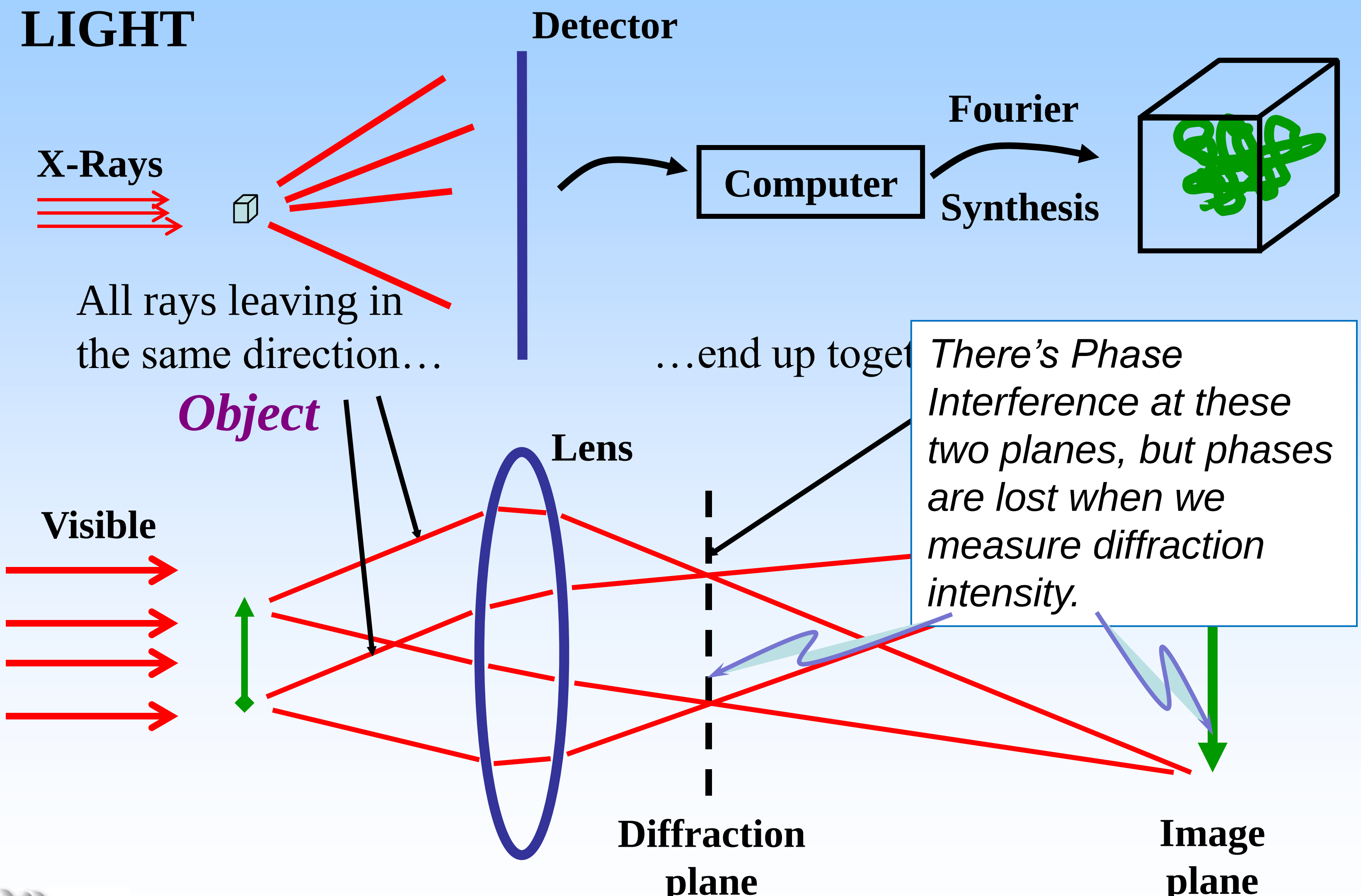
Creation of a molecule's image from a crystal has similarities to creating an image with a lens



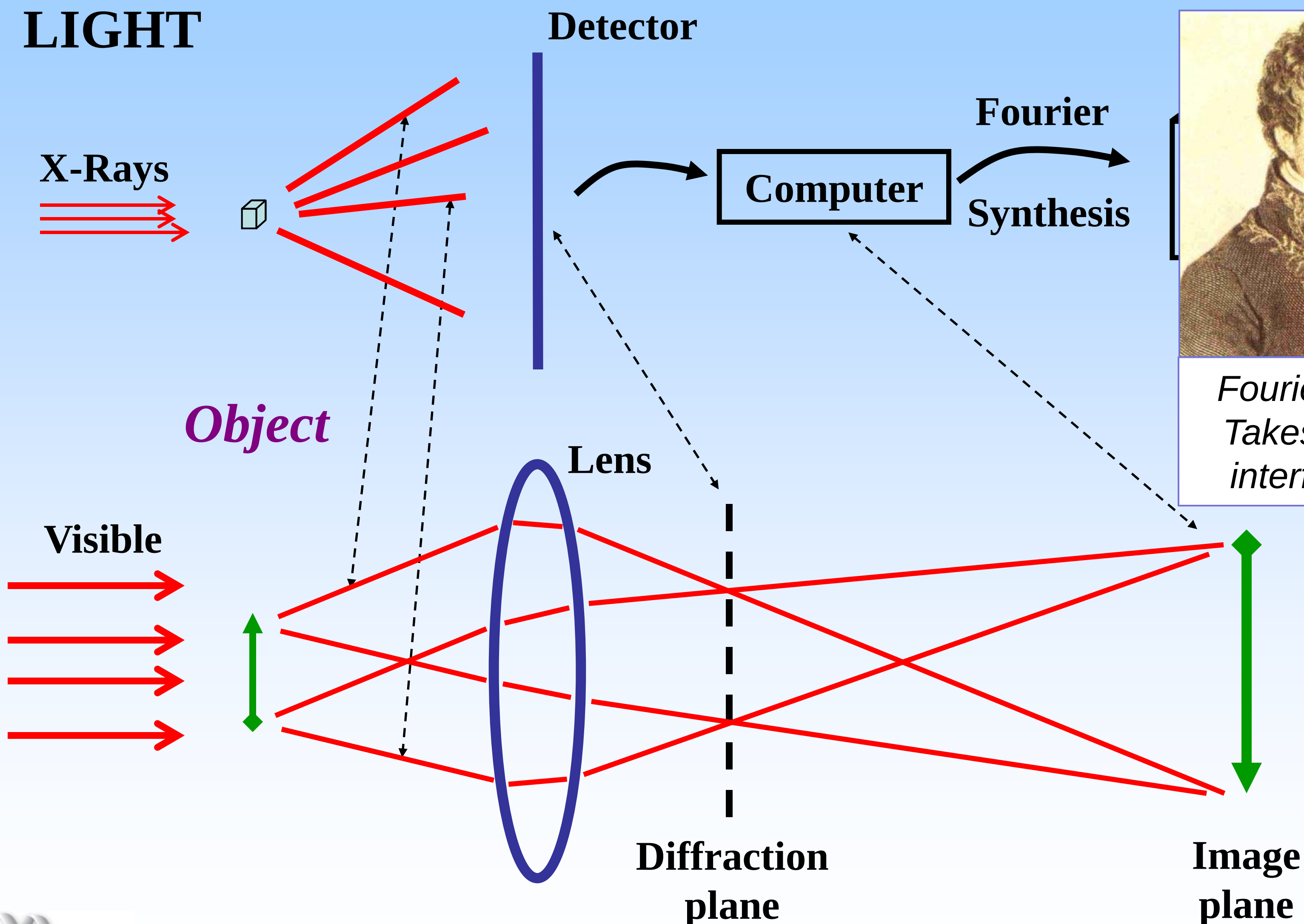
You already understand a little about how lenses work



Maybe you didn't know …



We use a crystal to give us diffraction, and computation to do the rest of the work of the lens.



Fourier Synth. Takes care of interference.

We'll see that the diffraction pattern gives information about the dimensions and periodicity of each view of the object.

Why do we use x-rays?

- The features we're trying to see are on the order of the **distance between atoms: 10^{-10}** meters.
- To "see" the atoms, we need to use light with a **wavelength** that is near to **this distance**.
- X-Rays (x-ray light) have a **suitable wavelength**.
- The x-rays are scattered by the electrons on the atoms so what we **see** is the **electrons**.

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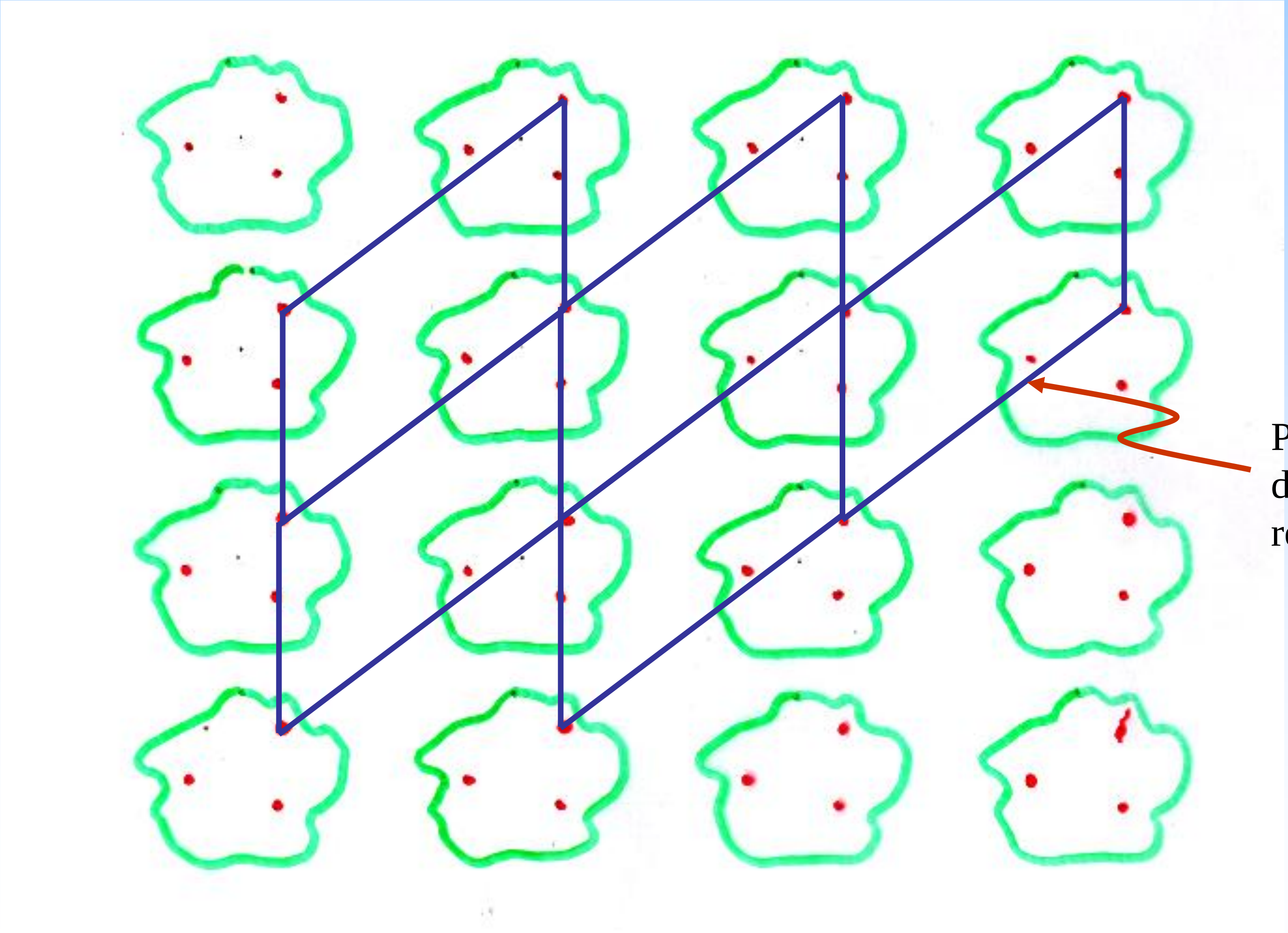
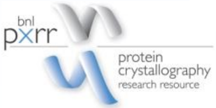
What is a **crystal**?

- A crystal is a **periodic arrangement** of objects (molecules) repeating in two or three dimensions.
- The **repeating unit** is a parallelepiped (in 3-D) or a parallelogram (in 2-D).
- A crystal of a typical protein will be half a mm on a side and contain **10^{15} molecules**.

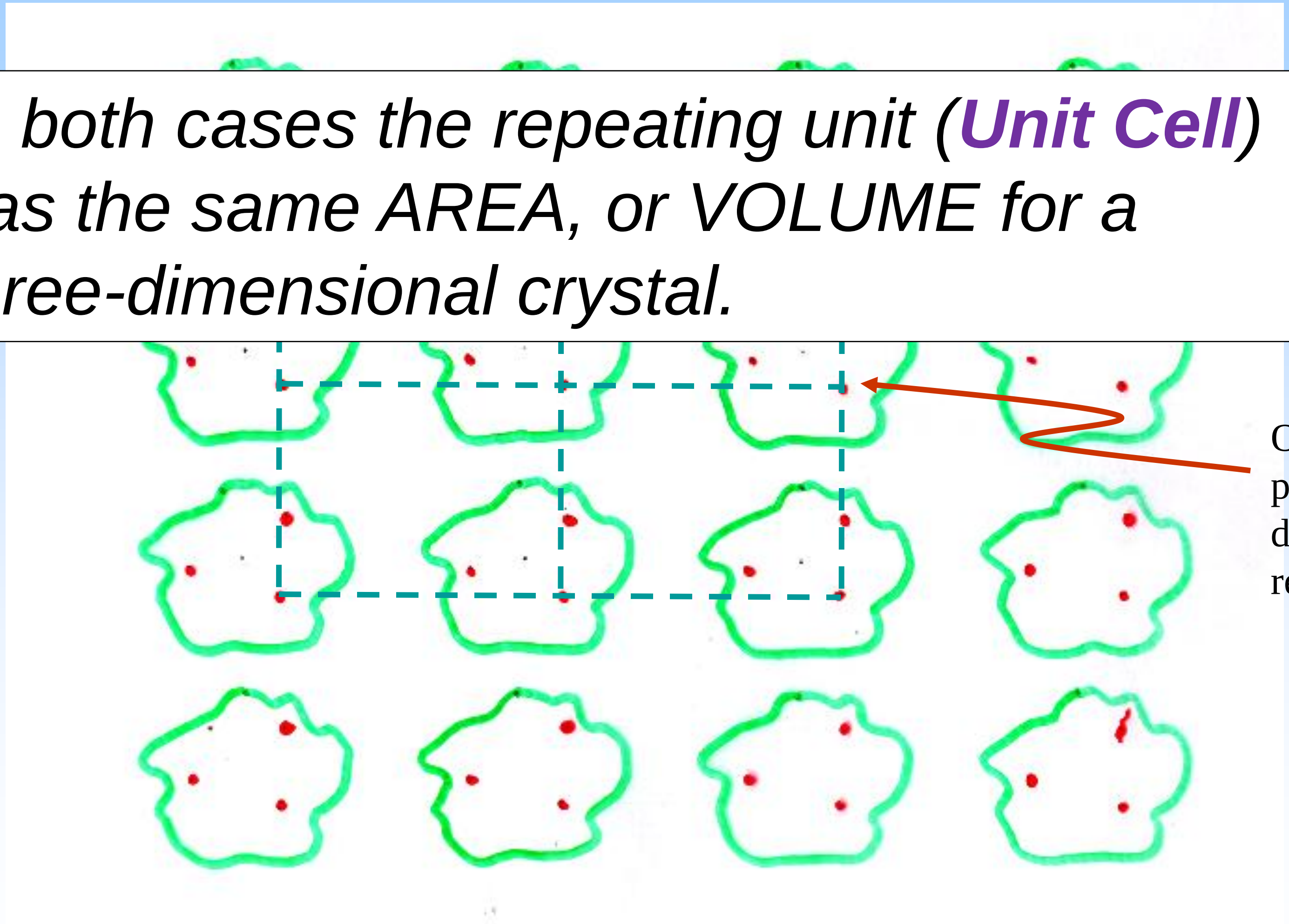
Here's one choice of repeating unit in this crystal made of apple trees



Parallelograms defining crystal repeat.



We could make a different choice of repeating unit



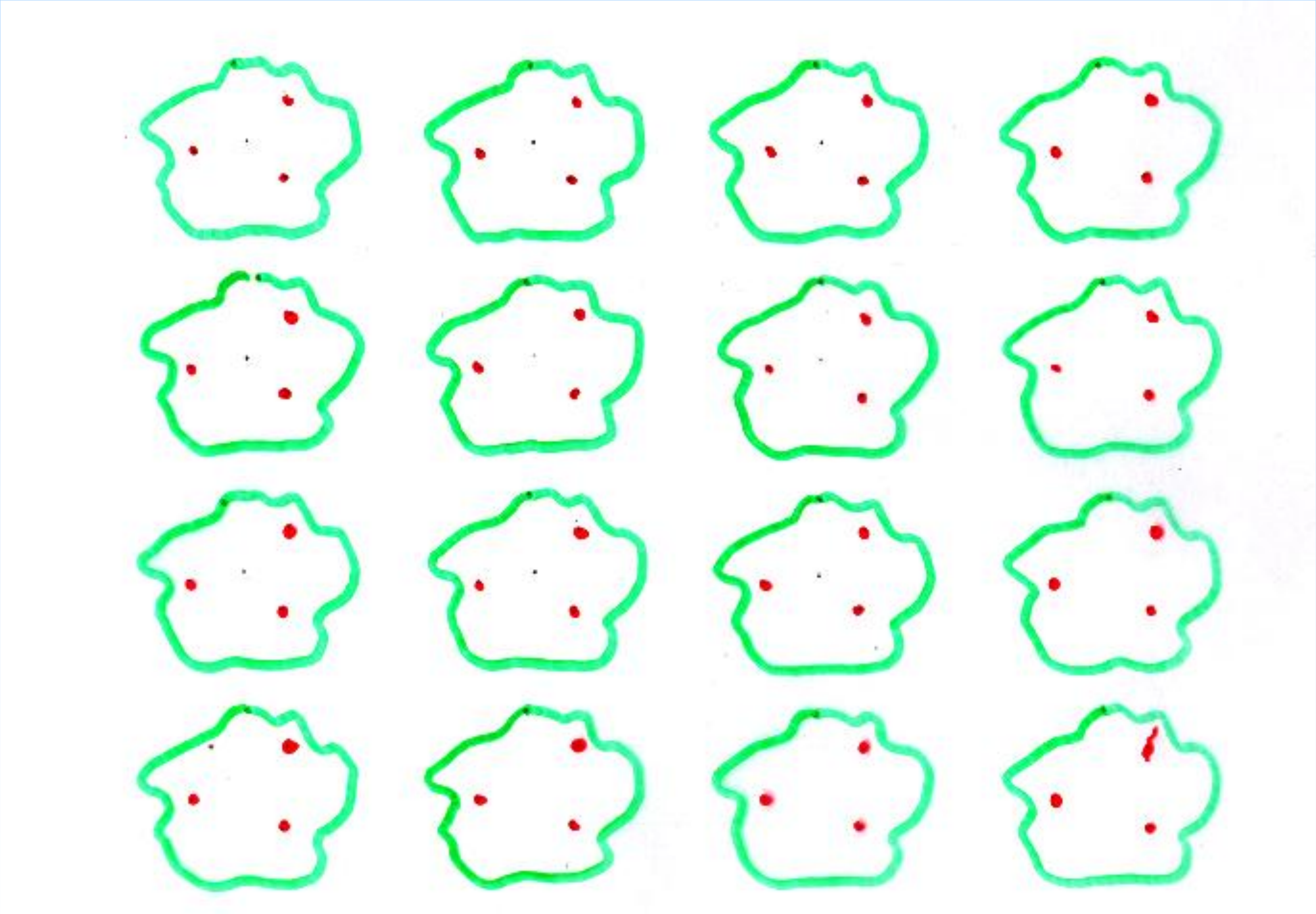
*In both cases the repeating unit (**Unit Cell**) has the same AREA, or VOLUME for a three-dimensional crystal.*

Other parallelograms defining crystal repeat.

Why do we use crystals when we'd like to see one molecule?

- We can't **focus** enough x-rays into a small enough volume to "see" a molecule. We use lots of molecules in a **crystal** to get a bigger target.
- Even if we could focus them, the x-rays would **burn up** the molecule.
- Even if that would work, we **don't have a lens** for the x-rays.
- The crystal **amplifies the signal**, and gives us a way to get the phase information back.

Let's return to our crystal made of apple trees, and define "planes" in that crystal.



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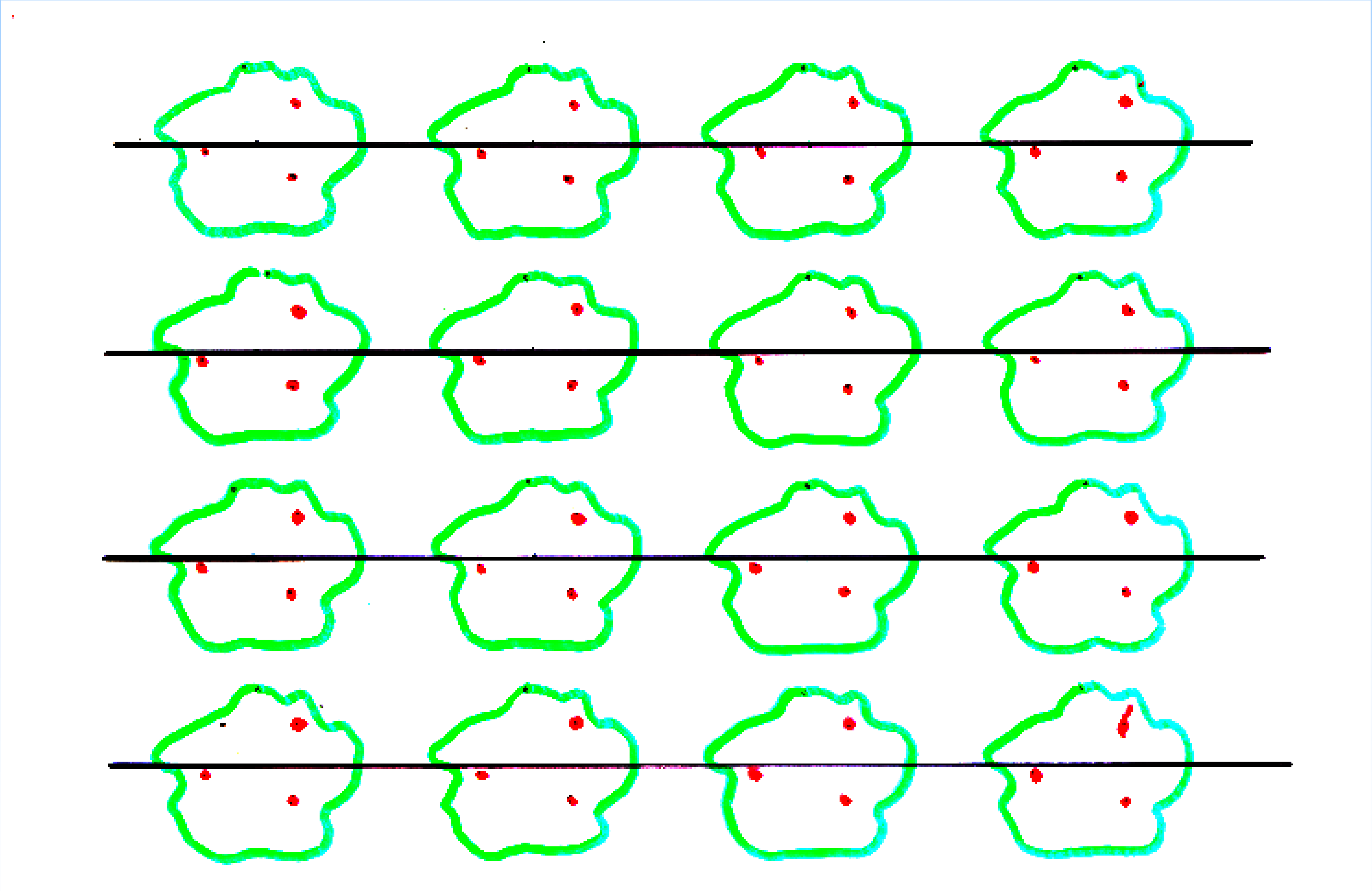
We can slice the crystal at lattice points: all planes pass through the same apple



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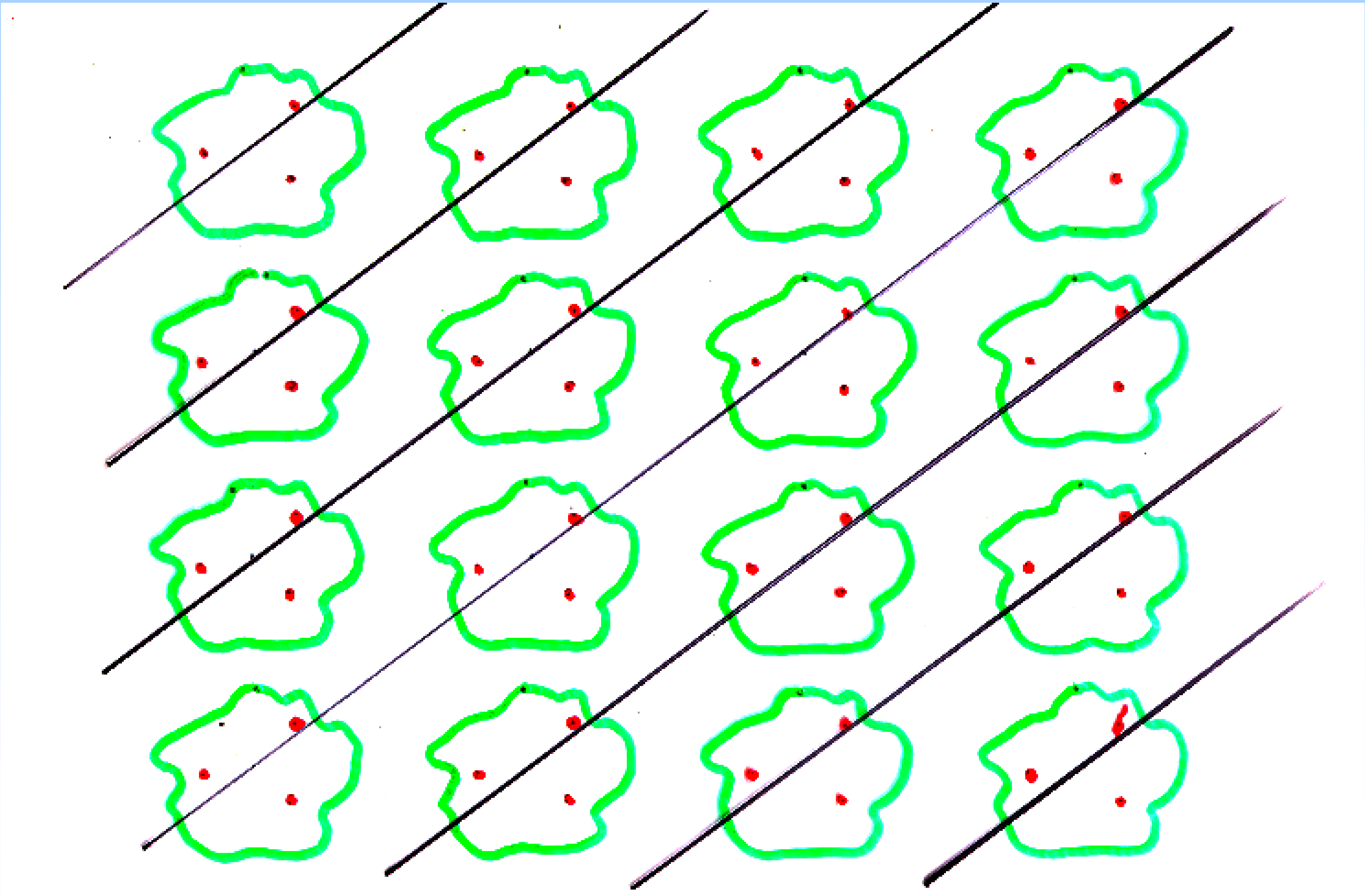
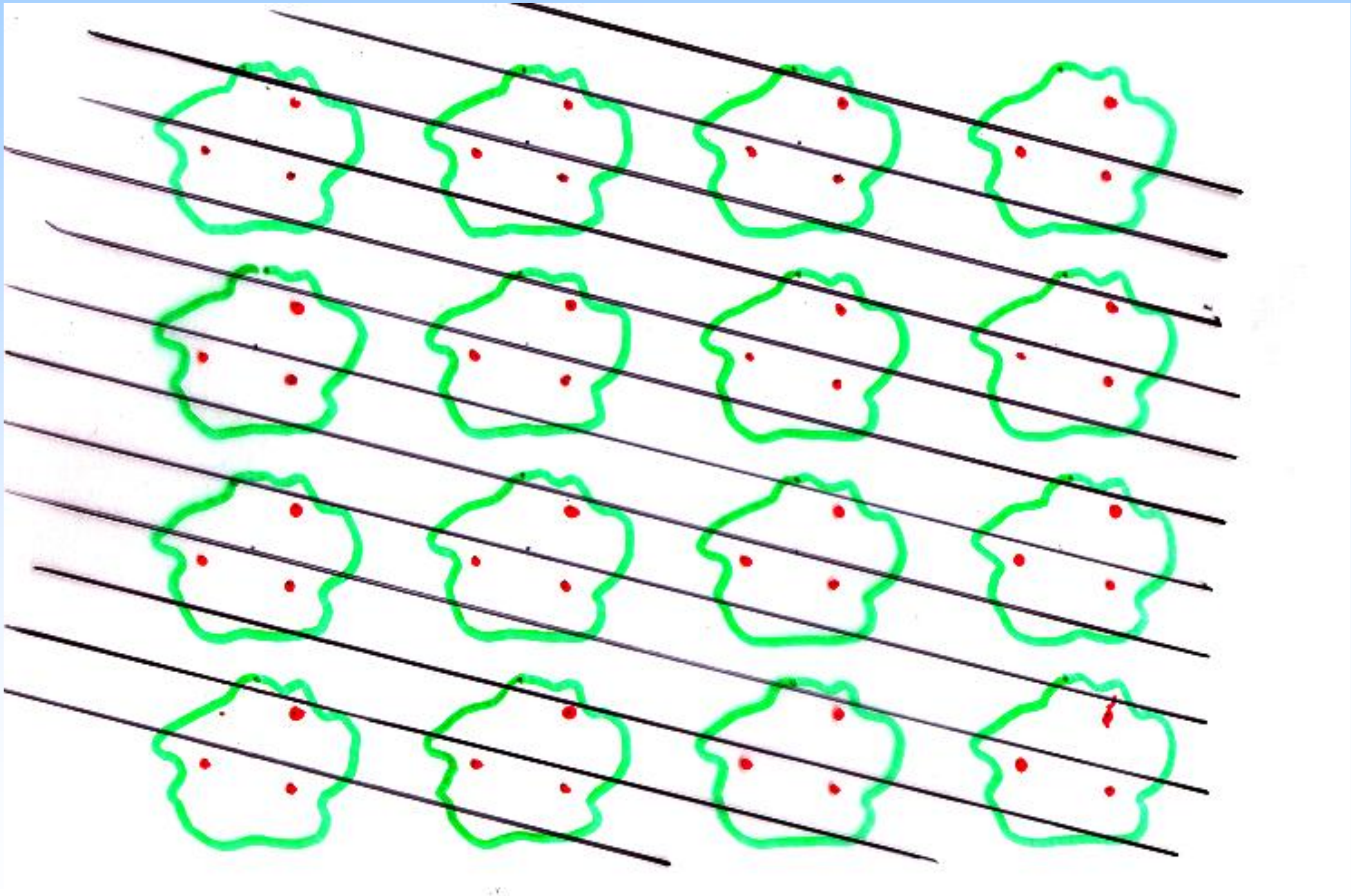
And at other angles. Notice:

- planes all pass by the same apple;
- the "stuff" between pairs of planes is always the same.



And one more time...



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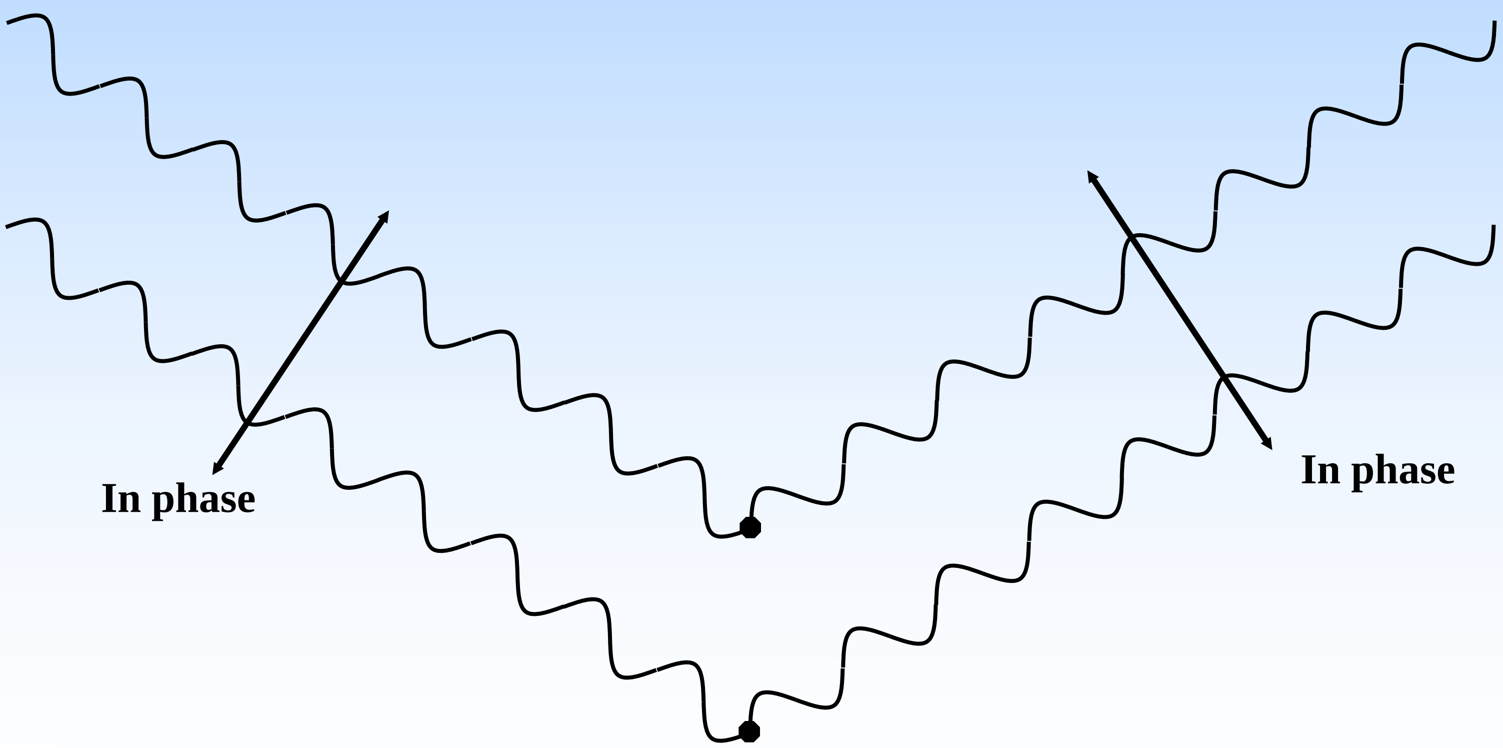
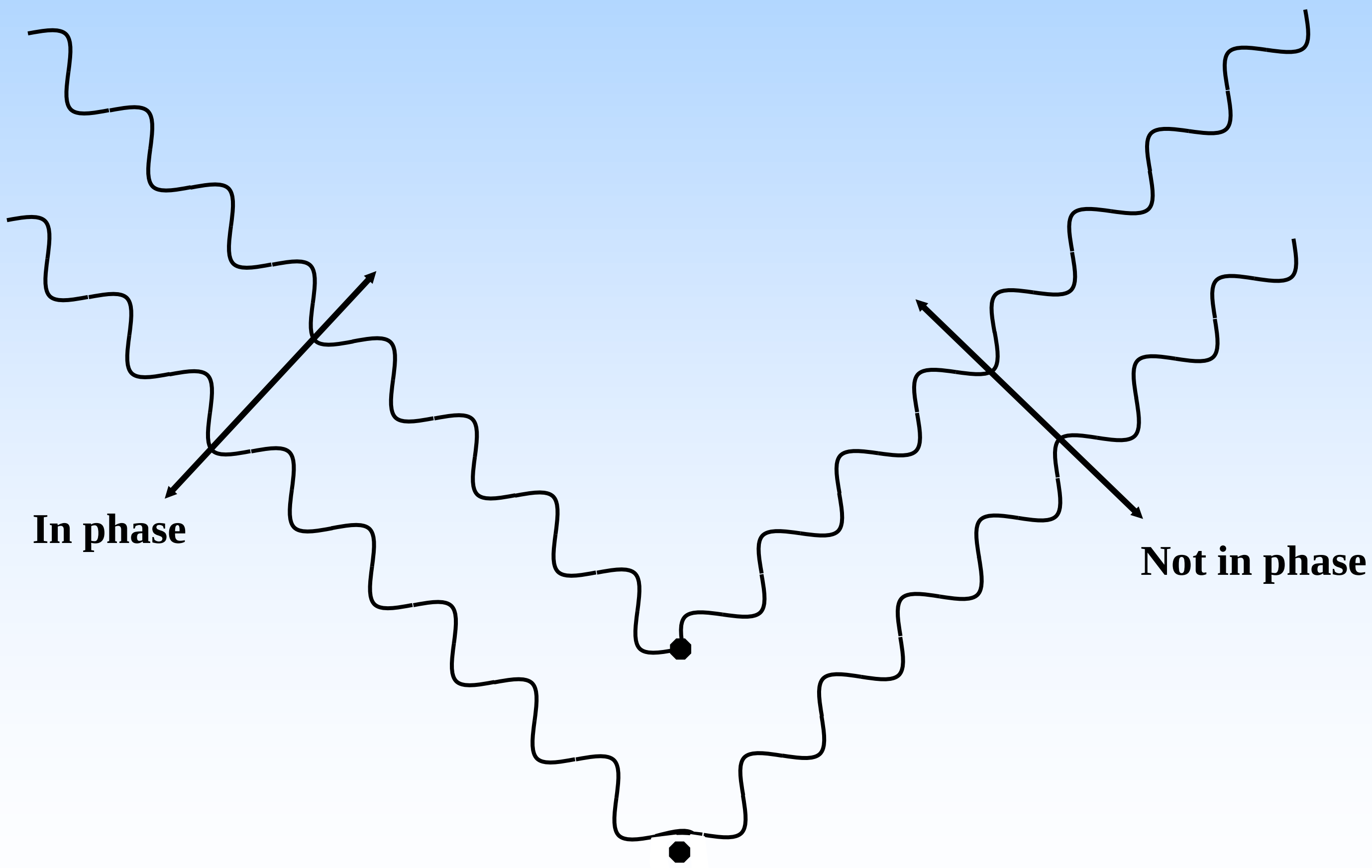
Diffraction – Let's do a thought experiment.

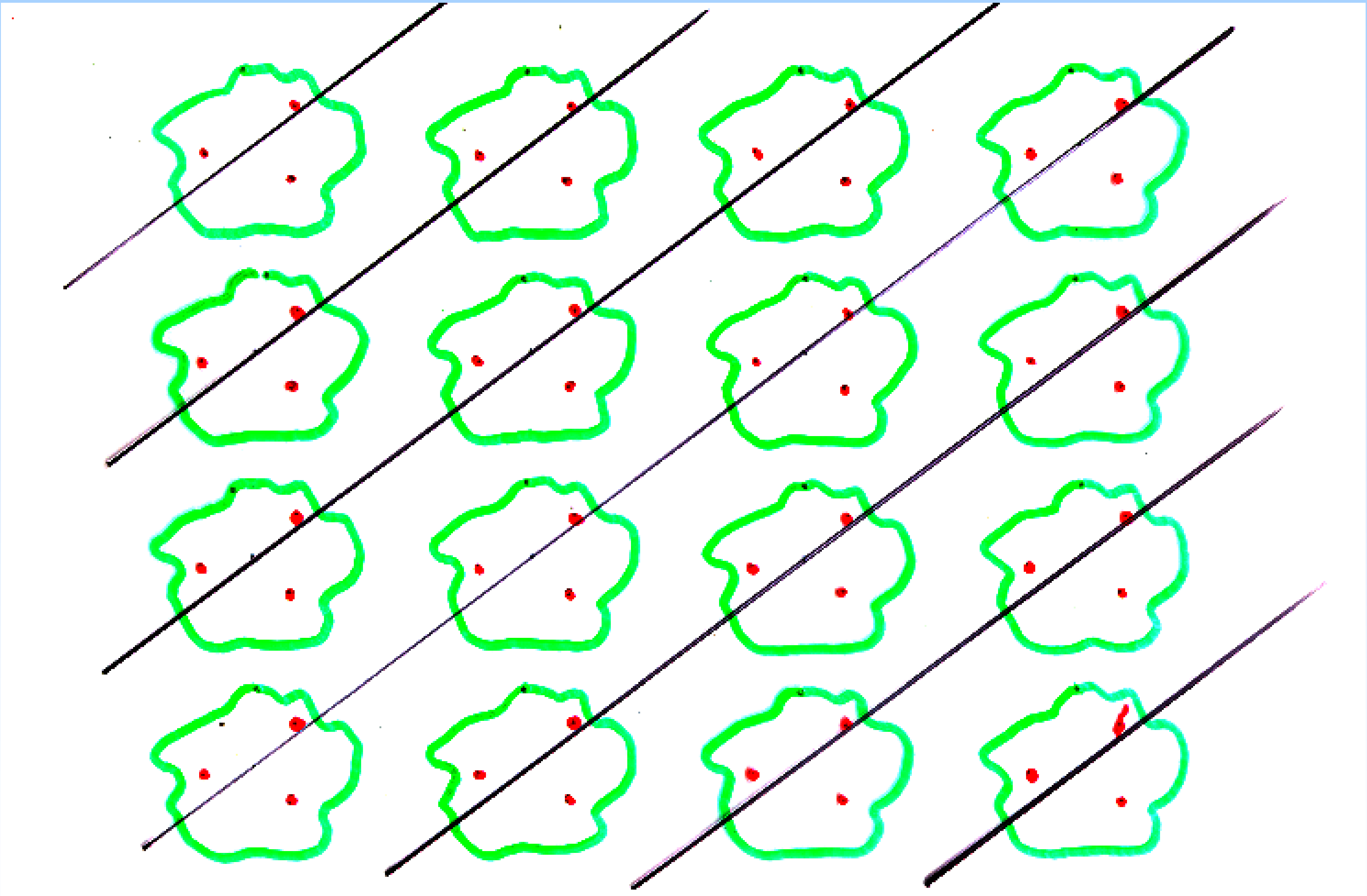
- Think of the material between the lattice planes as just two atoms, suspended in space.
- Send a beam of x-rays at these atoms.
- If the angle is just right for the wavelength and distance between the atoms, the scattered x-rays will be in phase, and they will interfere constructively.

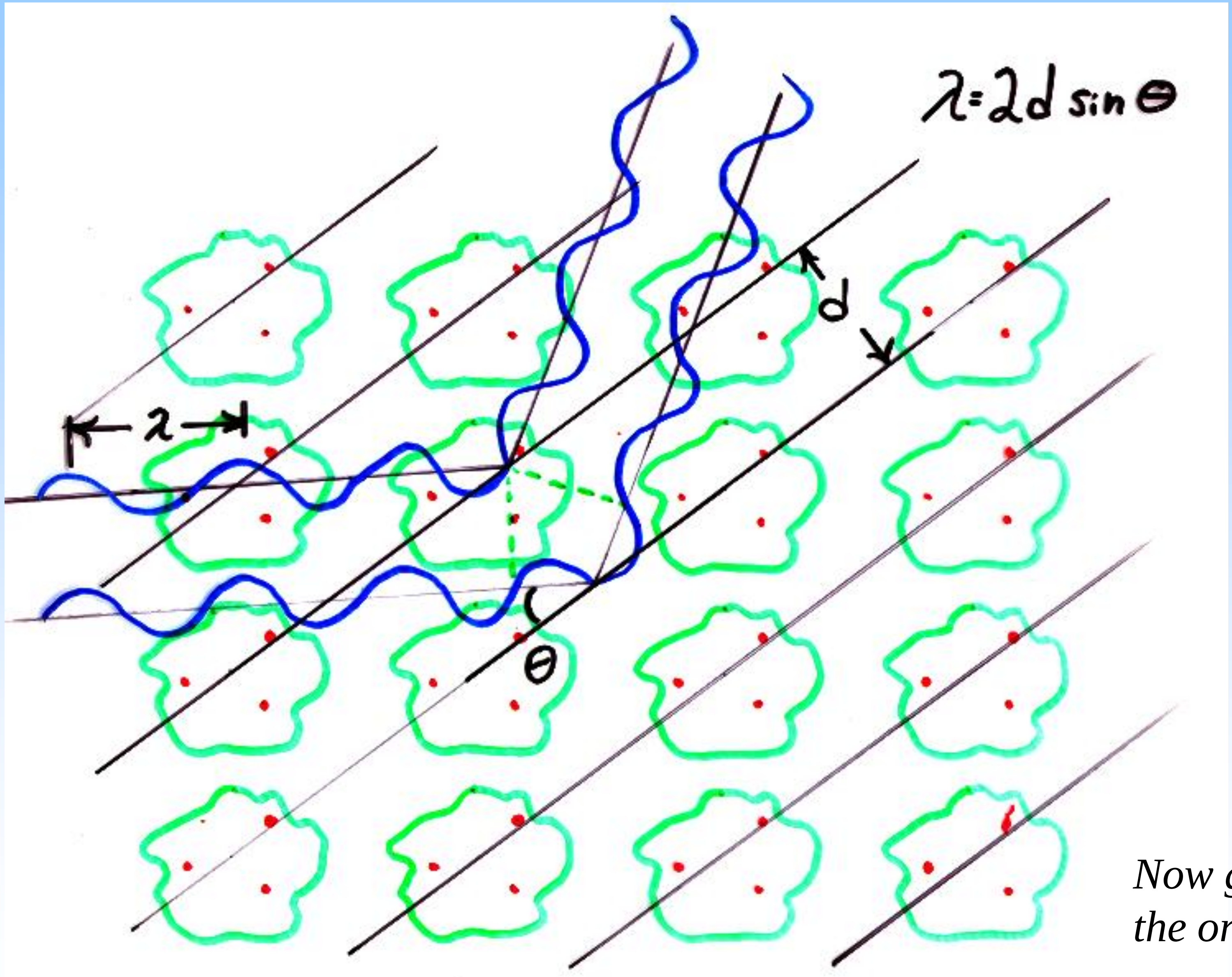


On the other hand, if things are not right, they won’t be in phase, and there will be no constructive interference, no diffraction.



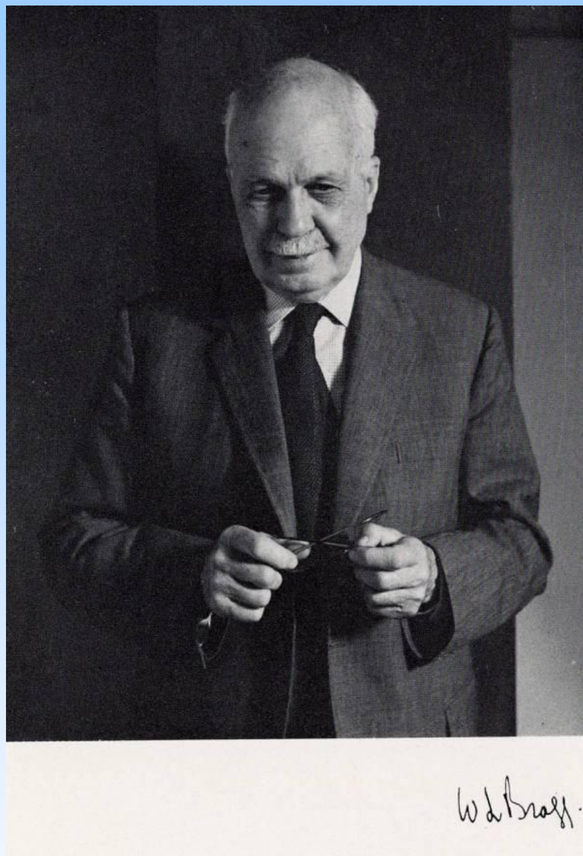
Now, as we think of the stuff between the lattice planes as being like each of those two atoms, we try to write a law that will show conditions to get diffraction.

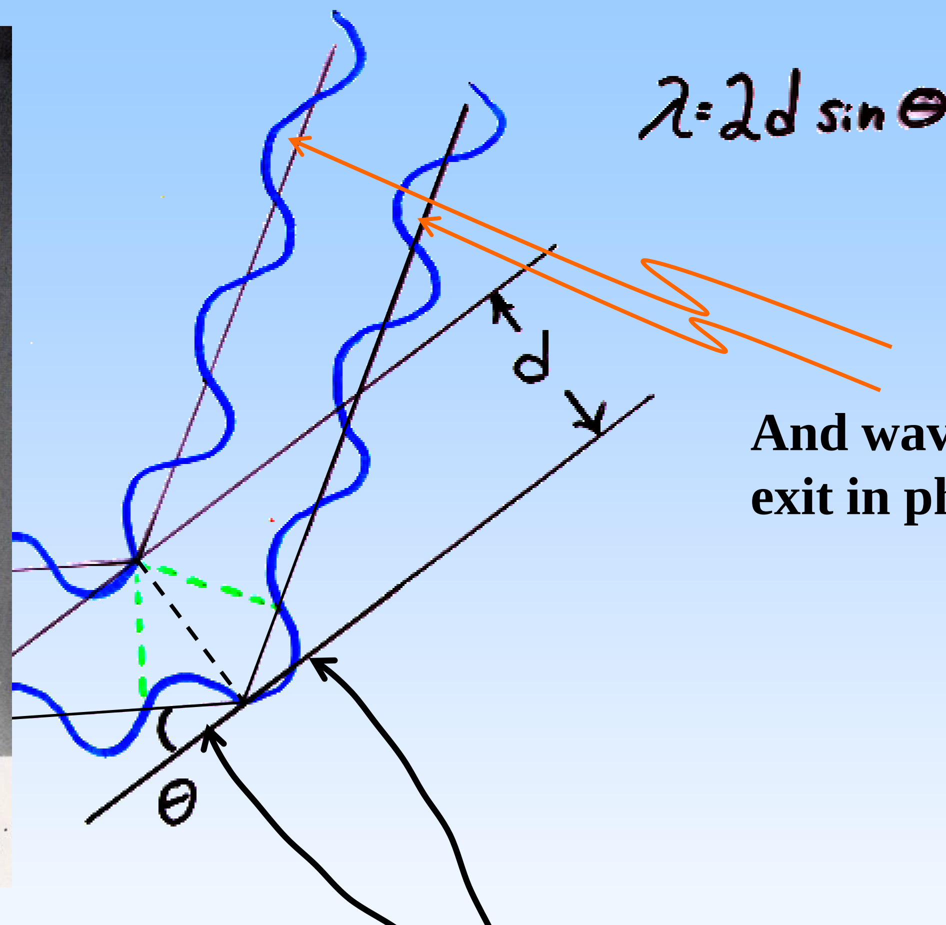




Now get rid of the orchard...

Bragg's Law describes diffraction as reflection from planes



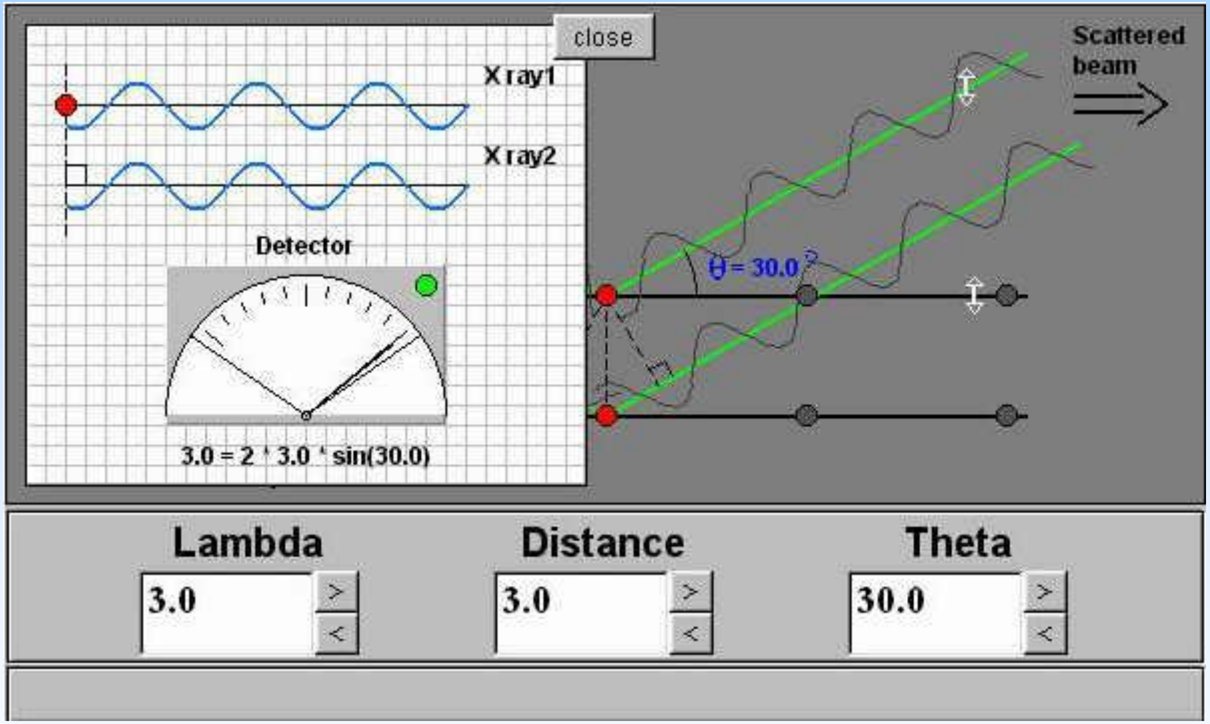


And waves exit in phase

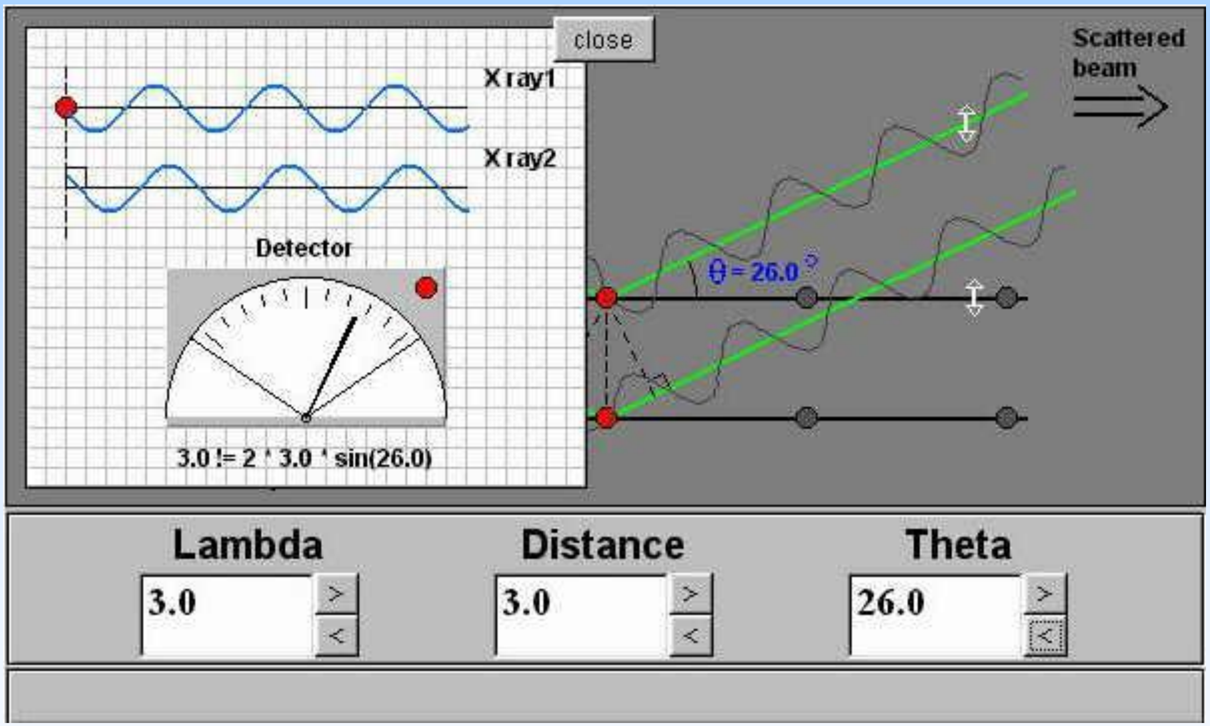
Waves come in "in phase."

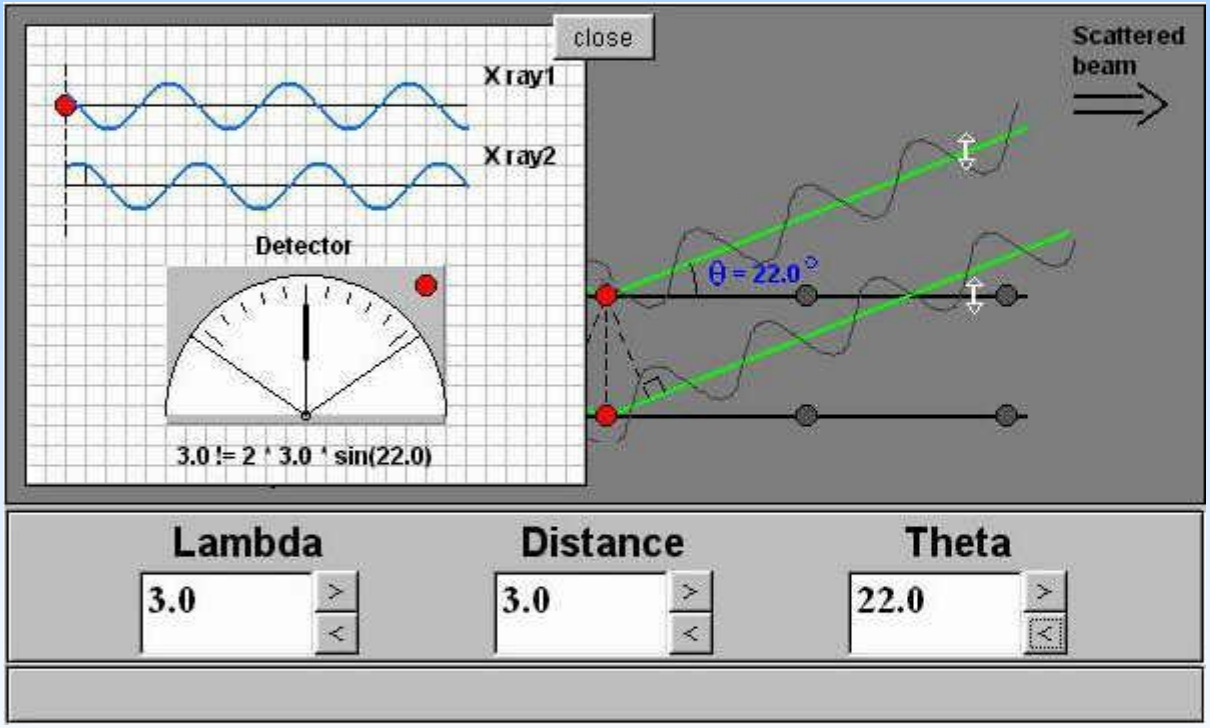
The wave travels exactly one wavelength to take the little detour

Watch what happens as we go from maximum to minimum diffracting position and back.

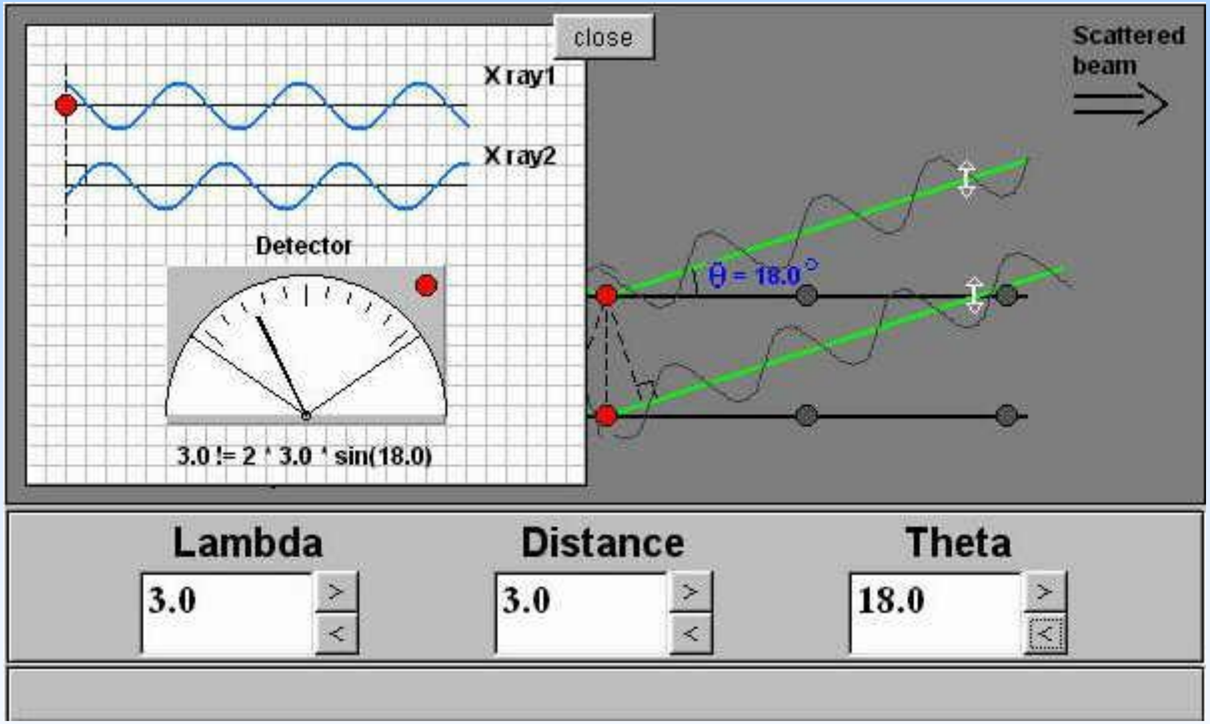


See: www.journeysunysbedu/ProjectJava/Bragg/home.html

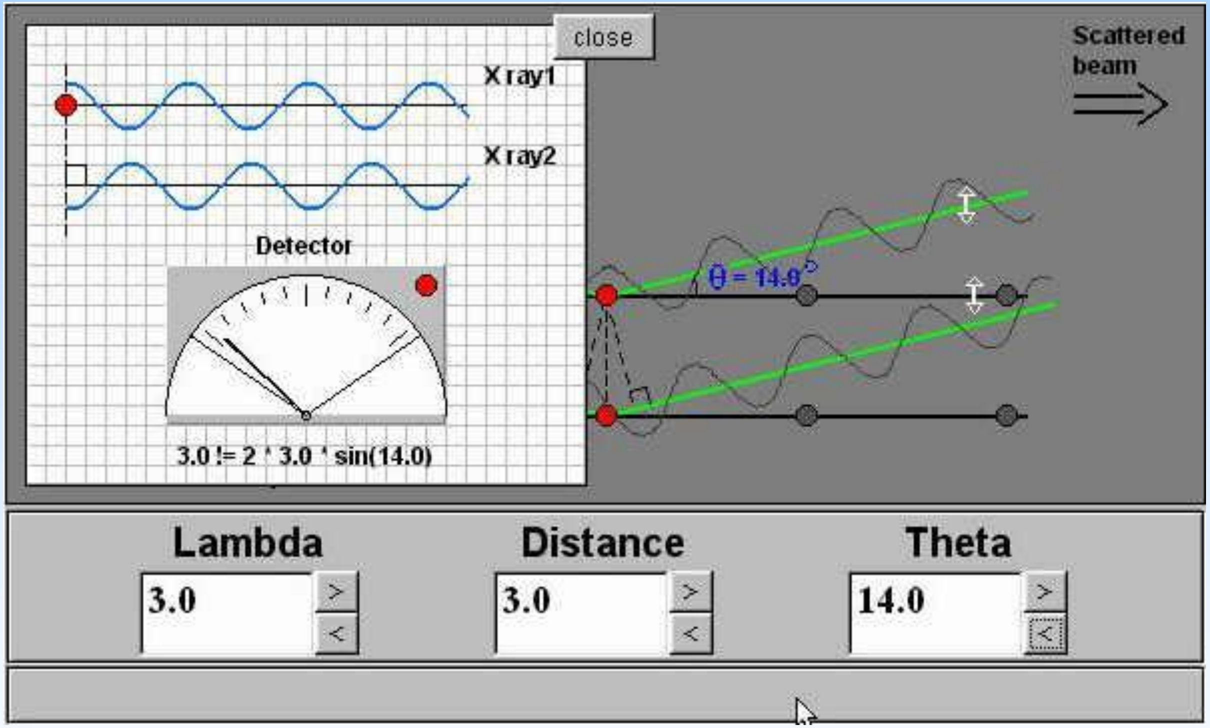

close
X ray1
X ray2
Detector
3.0 != 2 * 3.0 * sin(26.0)
θ = 26.0 °
Scattered beam
Lambda
3.0
Distance
3.0
Theta
26.0

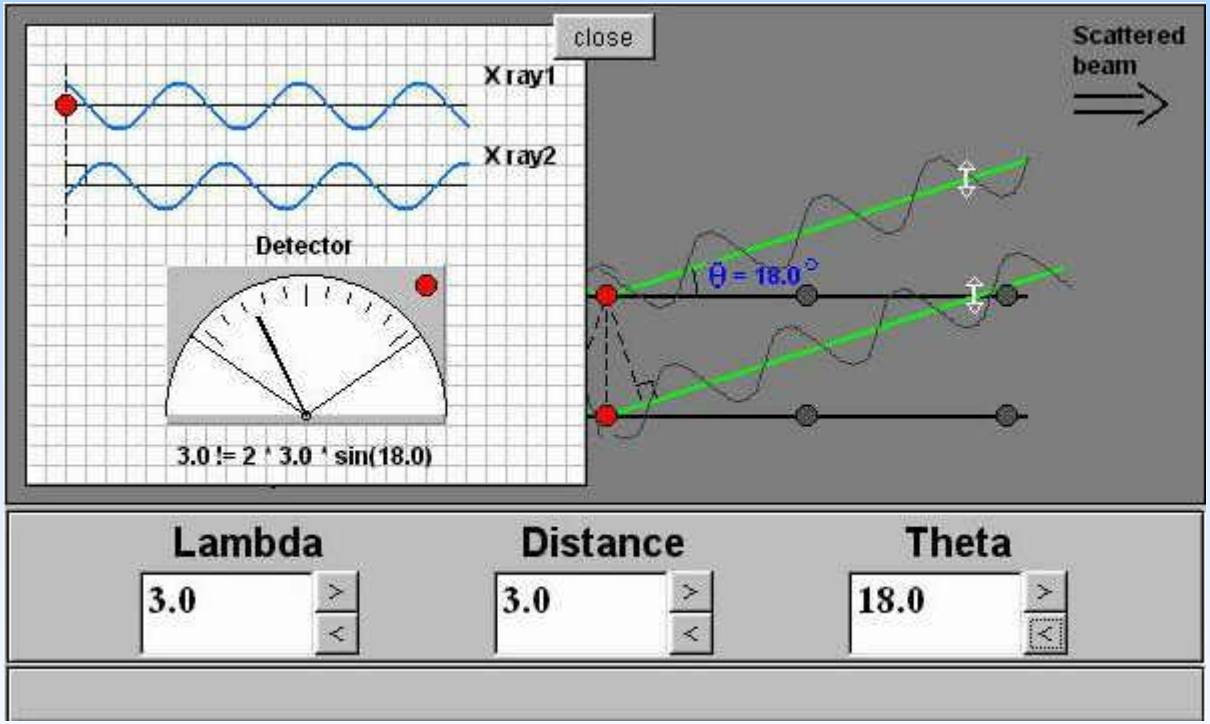


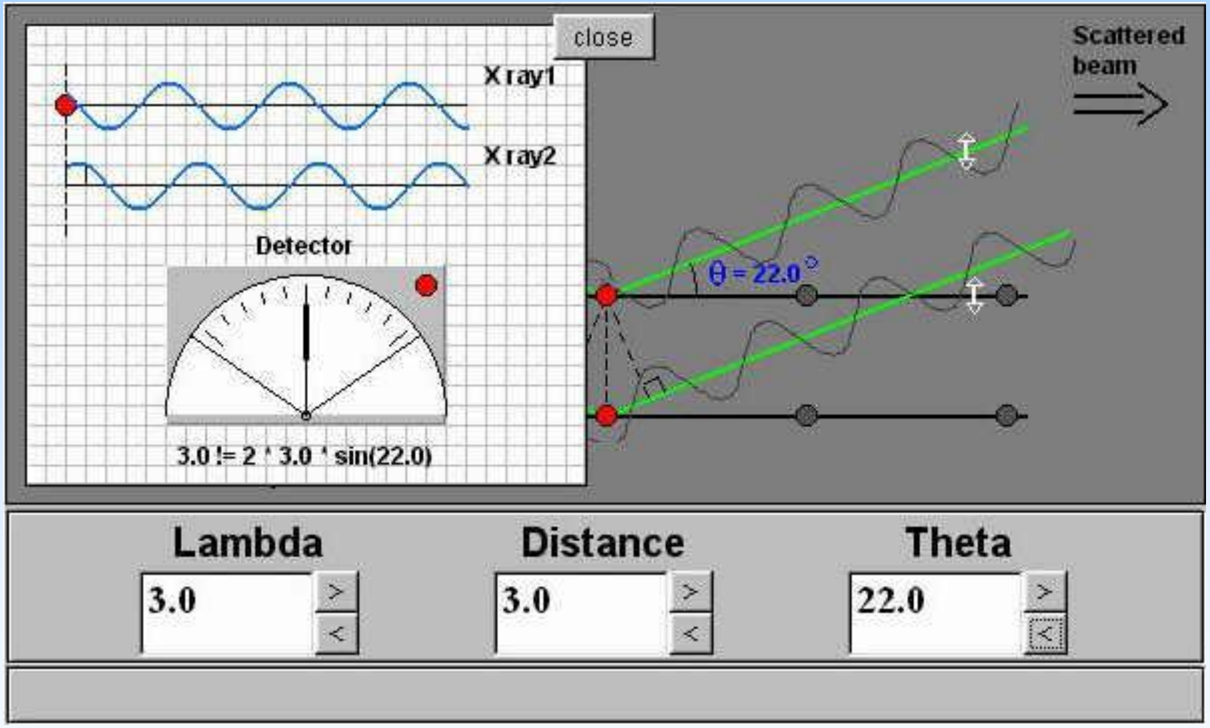
close
X ray1
X ray2
Detector
3.0 != 2 * 3.0 * sin(22.0)
Scattered beam
θ = 22.0°
Lambda
3.0
Distance
3.0
Theta
22.0

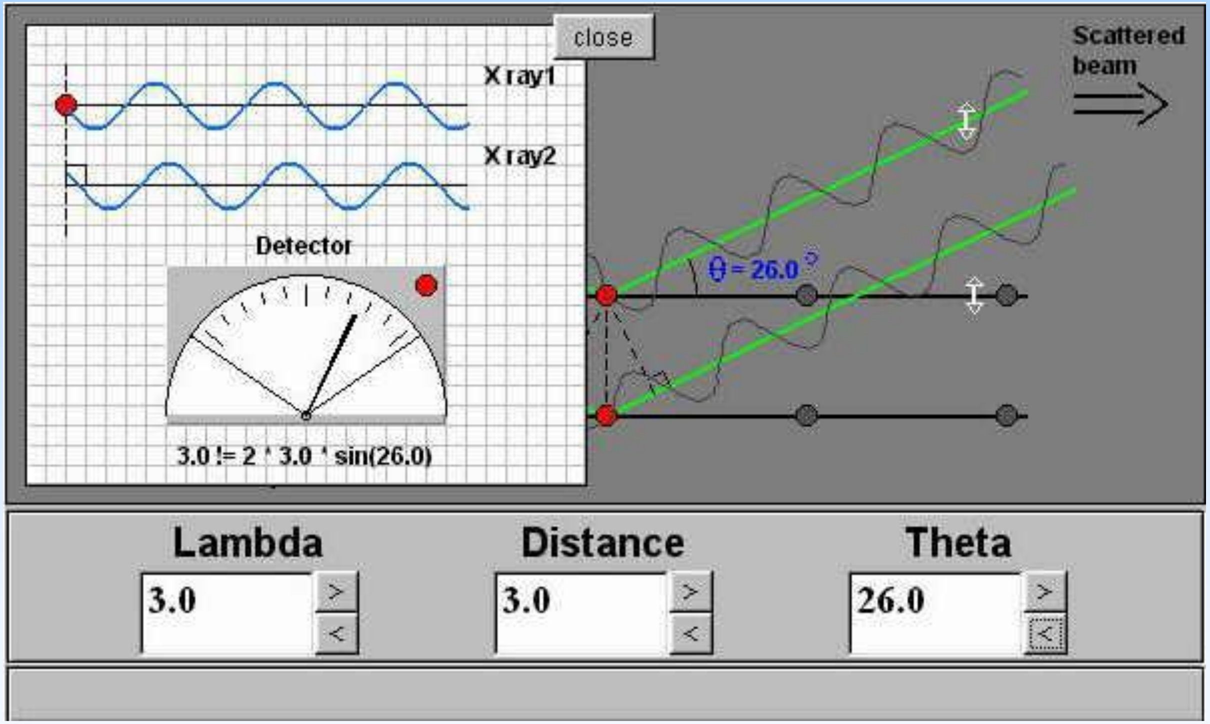


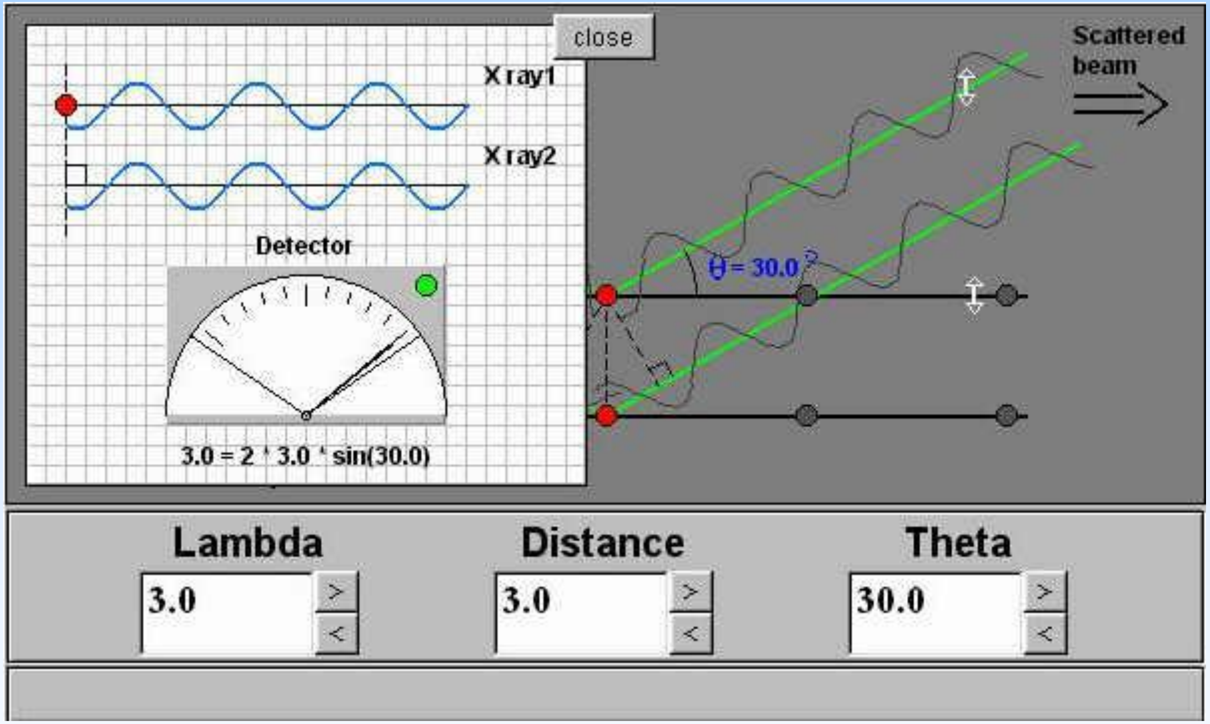
close
X ray1
X ray2
Detector
3.0 != 2 * 3.0 * sin(18.0)
Scattered beam
θ = 18.0°
Lambda
3.0
Distance
3.0
Theta
18.0


close
Scattered beam
X ray1
X ray2
Detector
θ = 14.0°
3.0 != 2 * 3.0 * sin(14.0)
Lambda
3.0
Distance
3.0
Theta
14.0


close
X ray1
X ray2
Detector
3.0 != 2 * 3.0 * sin(18.0)
Scattered beam
θ = 18.0°
Lambda
3.0
Distance
3.0
Theta
18.0

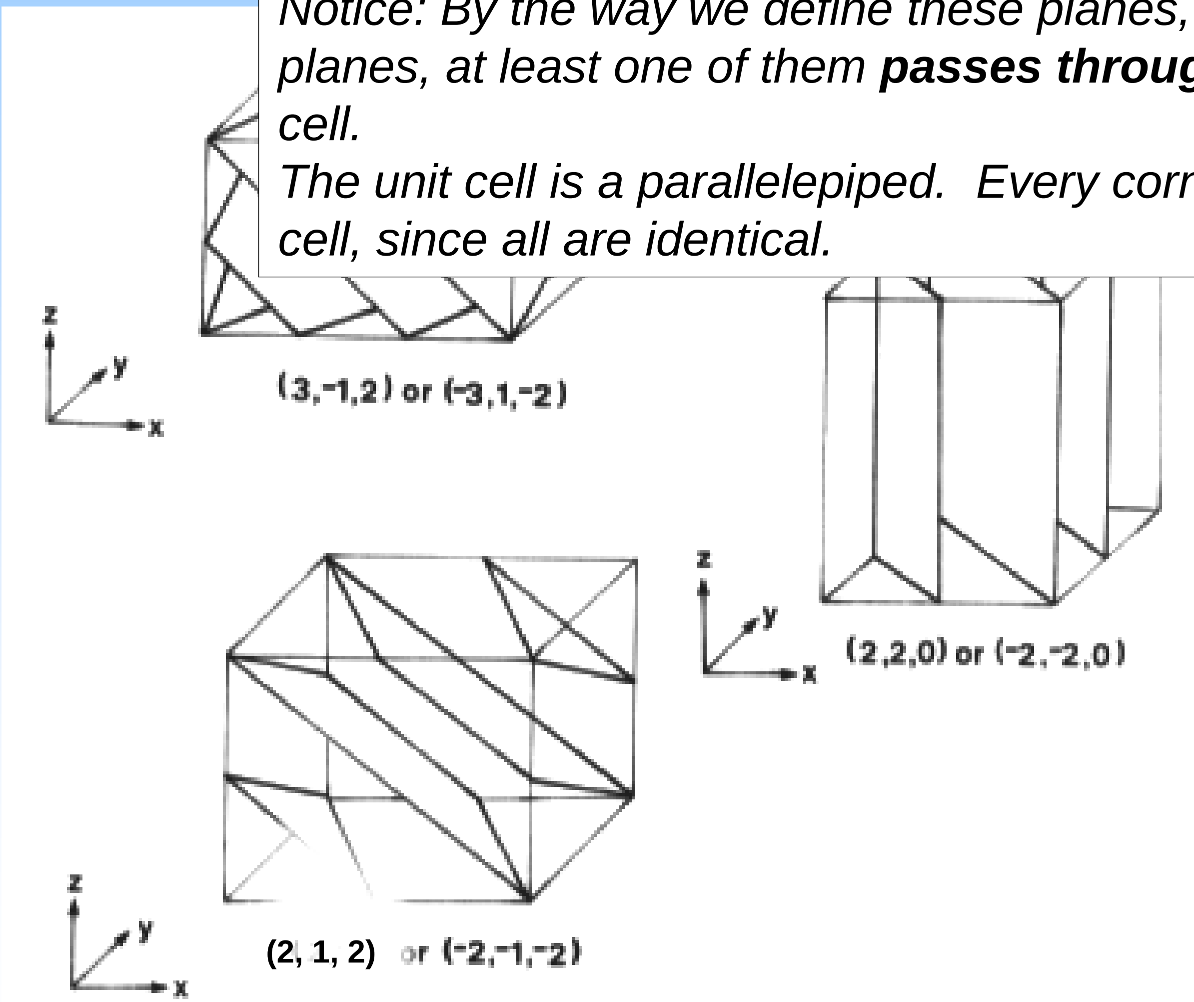

close
X ray1
X ray2
Scattered beam
Detector
θ = 22.0°
3.0 != 2 * 3.0 * sin(22.0)
Lambda
3.0
Distance
3.0
Theta
22.0


close
X ray1
X ray2
Detector
3.0 != 2 * 3.0 * sin(26.0)
θ = 26.0 °
Scattered beam
Lambda
3.0
Distance
3.0
Theta
26.0


close
X ray1
X ray2
Detector
3.0 = 2 * 3.0 * sin(30.0)
Scattered beam
θ = 30.0 °
Lambda
3.0
Distance
3.0
Theta
30.0

We have a way to "index" planes in a paralellepiped to give a unique description of them.

*Notice: By the way we define these planes, for every family of planes, at least one of them **passes through the origin** of the unit cell.*
The unit cell is a parallelepiped. Every corner is an origin of a unit cell, since all are identical.



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pieces into which each plane cuts the axis of the "unit cell" of the crystal -- the smallest repeating unit that makes up the crystal.

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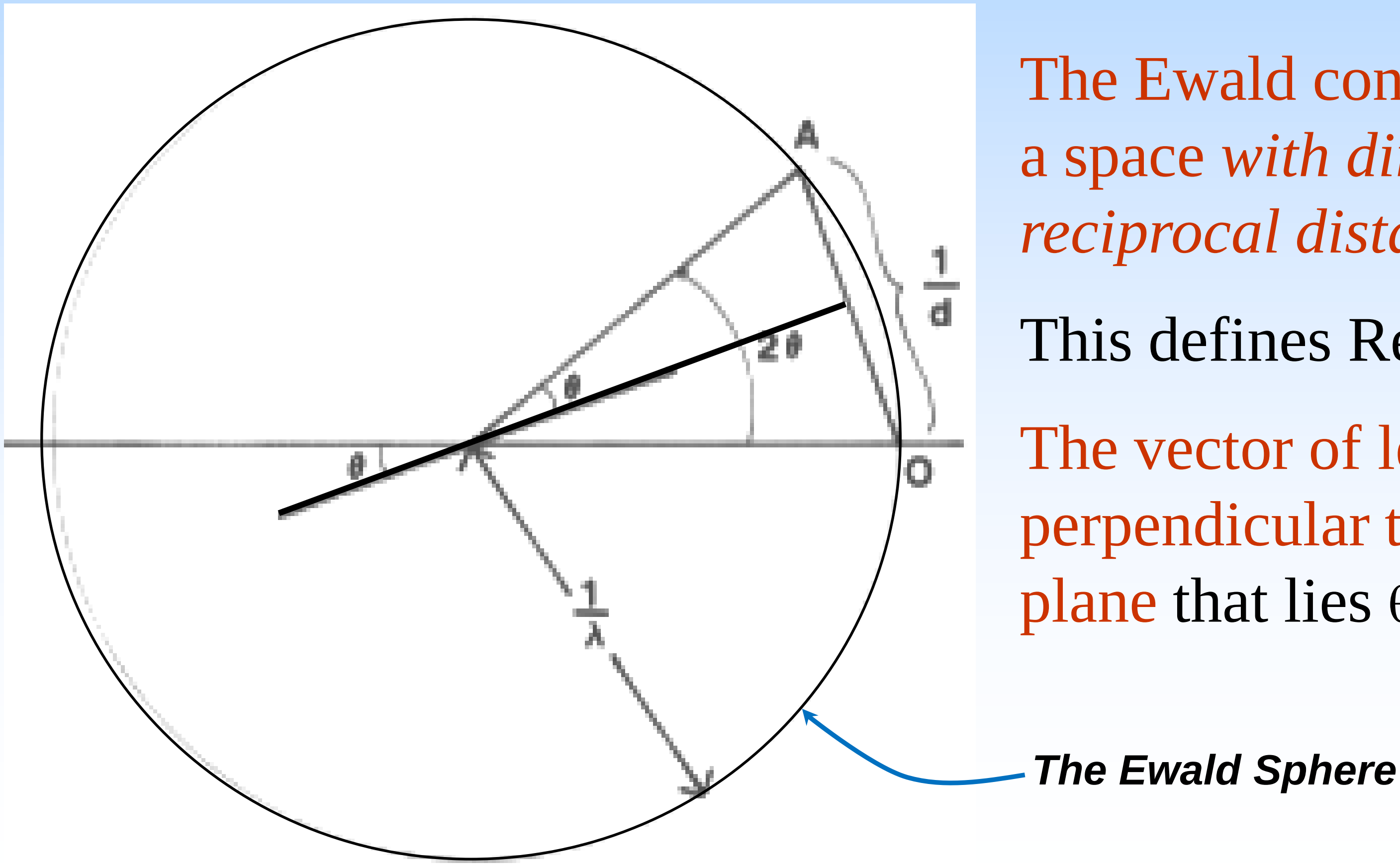
To relate the planes in the crystal lattice to the points in the diffraction pattern, we make Ewald's construction.

We have that $\sin\theta = (OA/2)/(1/\lambda) = \lambda \times OA/2$, or $\lambda = 2 \sin\theta/OA$.

Compare this to Bragg's Law: $\lambda = 2d \sin\theta$.

We take **1/OA as being equivalent to d**.

Notice the reflection plane, and that OA is perpendicular to it.

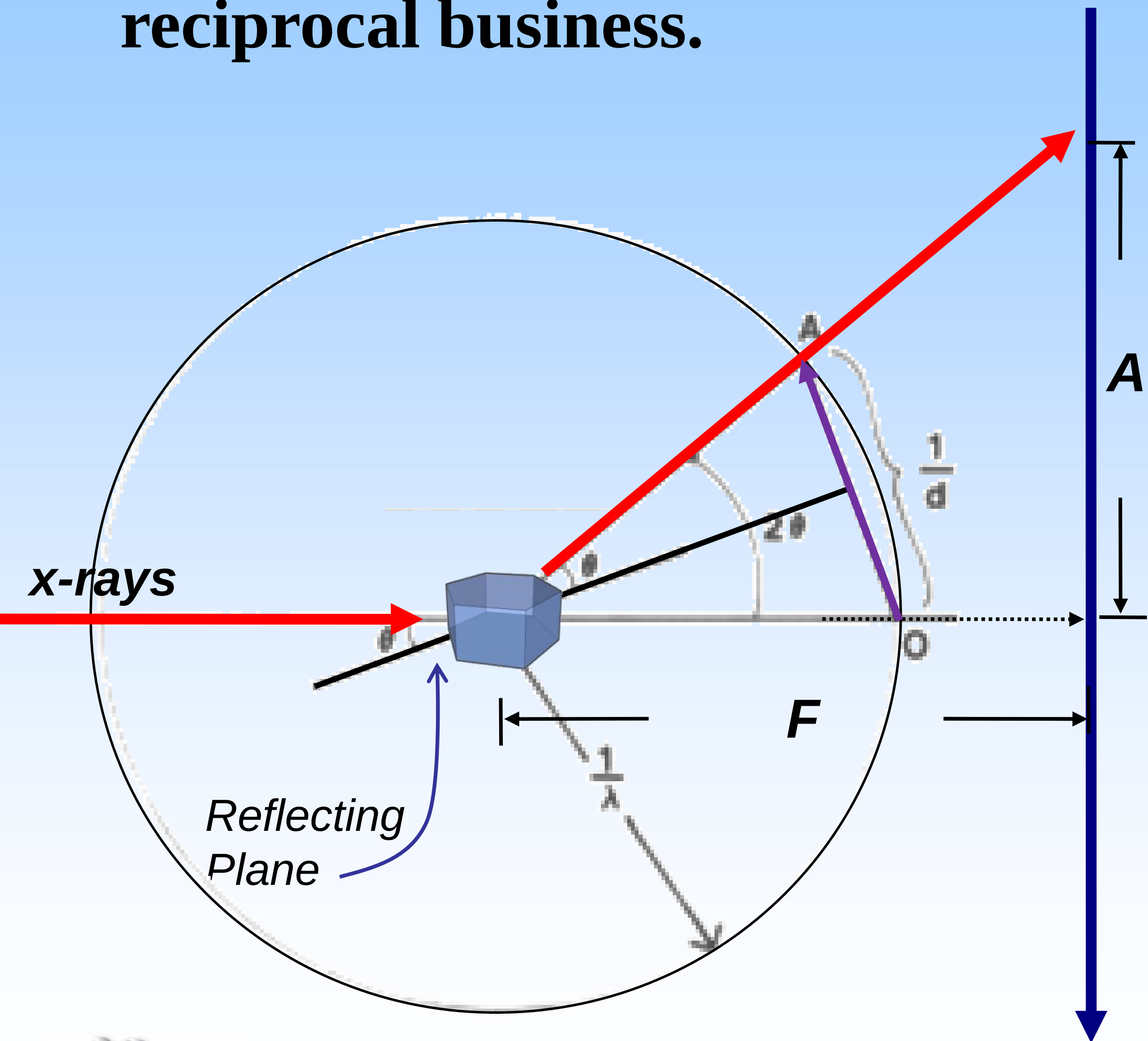


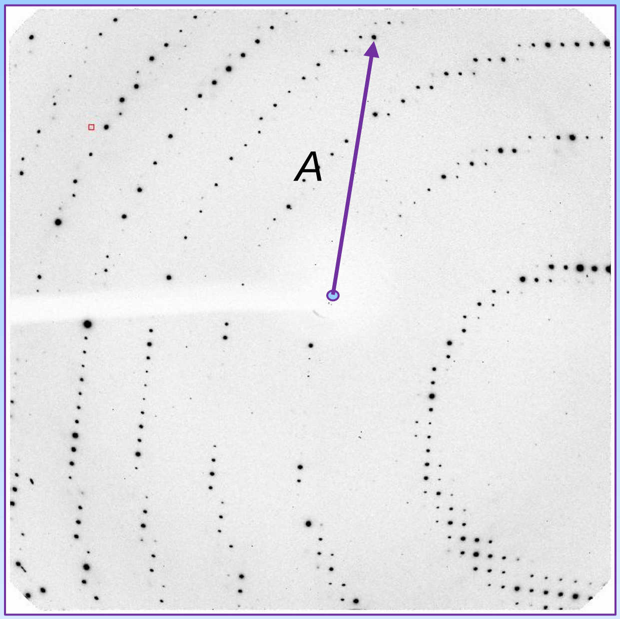
The Ewald construction exists in a space *with dimensions of reciprocal distance!*

This defines Reciprocal Space!

The vector of length 1/d is perpendicular to the reflecting plane that lies θ from the "rays."

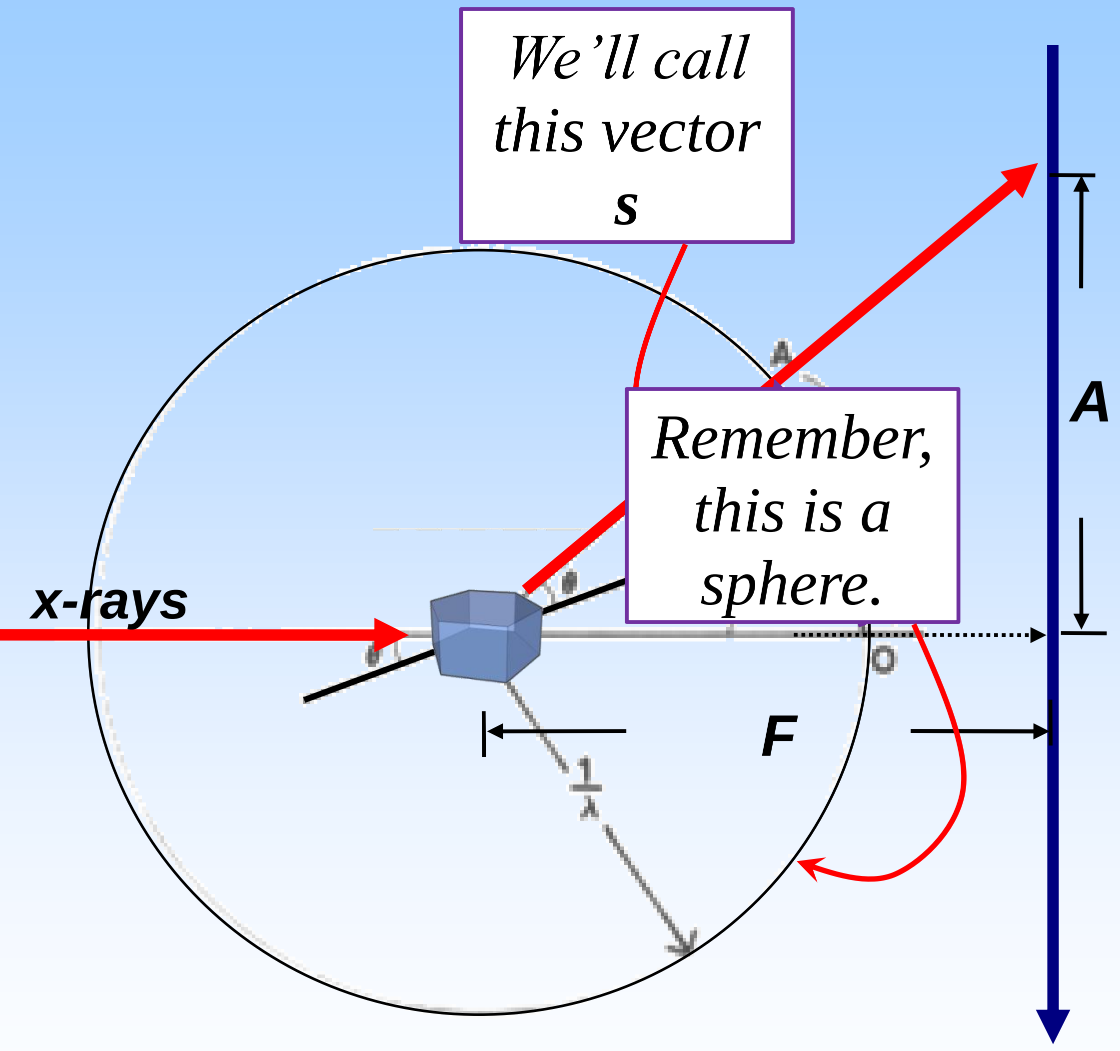
First, let's understand what's happening in the real experiment, then we'll try to understand the reciprocal business.

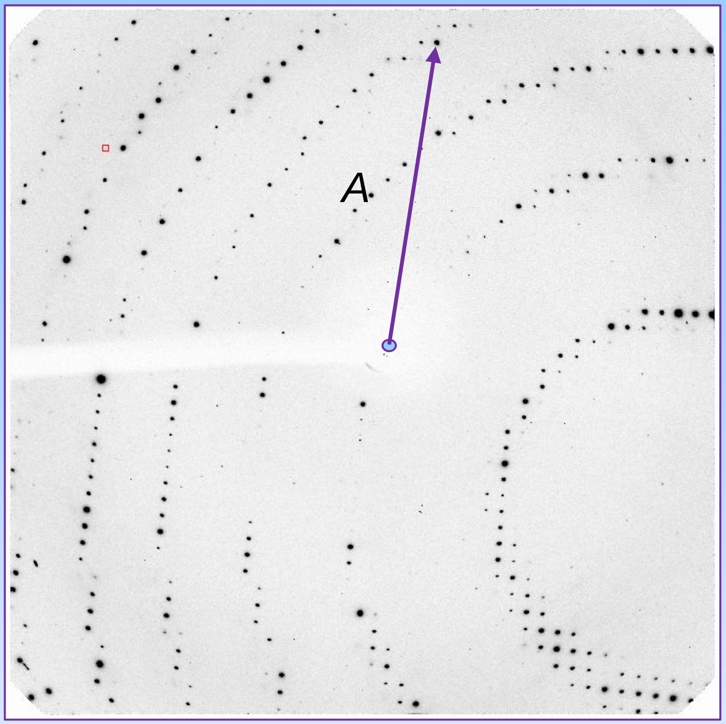




Bragg's Law is obeyed: diffraction occurs when a vector of length $1/d_{hkl}$, which is perpendicular to the lattice planes (hkl), ***touches the Ewald sphere*** of radius $1/\lambda$.

A little trigonometry:





A / F = tan(2θ)

λ = 2d sin(θ)

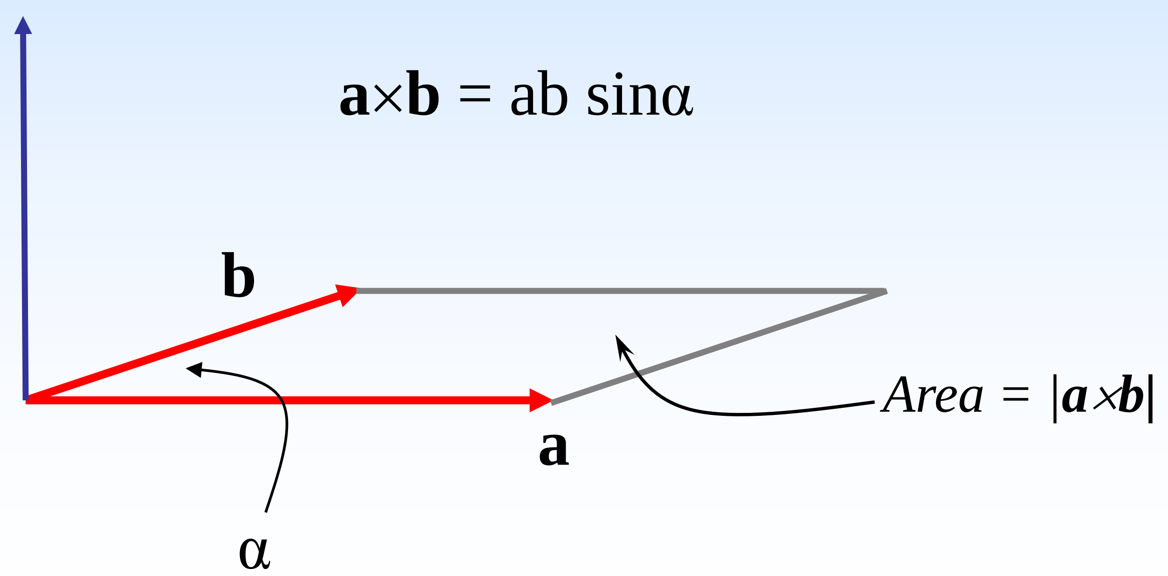
We can get the d-spacing for the reflection.

How can we define this vector that is perpendicular to the Bragg plane, and has a length that is the reciprocal of the distance between the planes?

We'll define the edges of a unit cell with three vectors. Start with **a** and **b**. We know that the cross product of two vectors lies **perpendicular** to the plane of the two vectors.

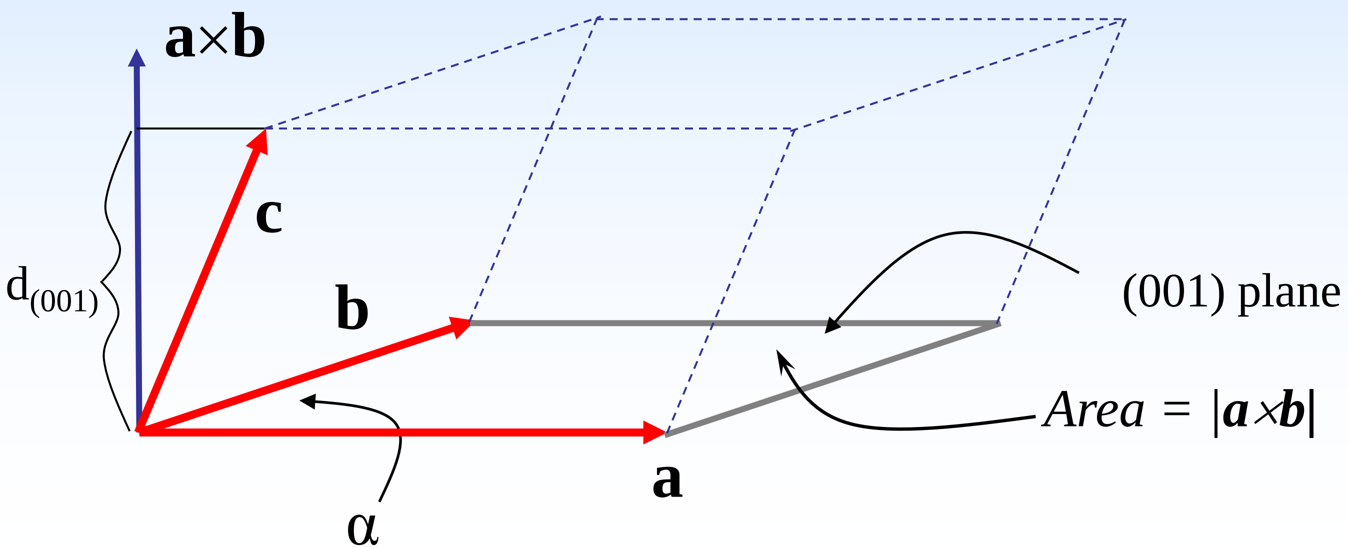
This is the direction we want. The **amplitude** of **a** **b** is the **area** of the parallelogram defined by the vectors:

$$\mathbf{a}\times\mathbf{b} = ab\sin\alpha$$



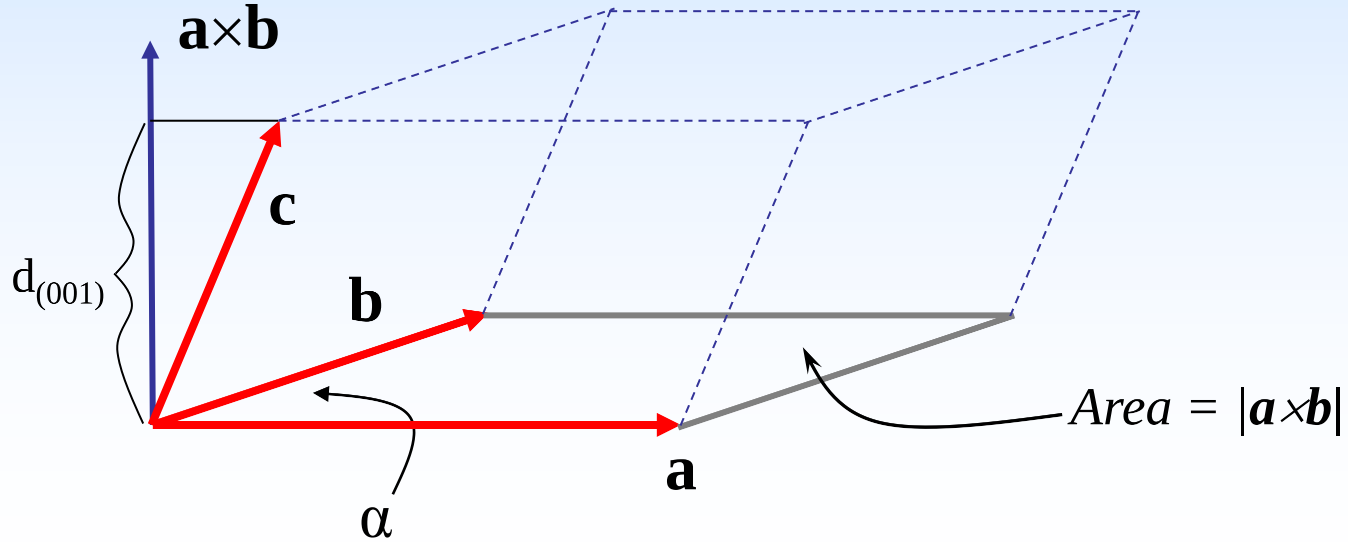
We've described the base of the unit cell of the crystal by two vectors a and b, and **the area** of the base is the **amplitude** of the cross product of **a** and **b**.

Now we'll include the third vector c. We want to know the spacing $d_{(001)}$, between the **ab** planes [the **(001)** lattice planes]. It must be the projection of **c** on the vector **a**×**b**. We know that we get the **product of the projection of one vector on another** with the vector dot product: $(\mathbf{a}\times\mathbf{b})\cdot\mathbf{c}$.



So **a×b·c**, known as a **vector triple product,** is the area of **ab** times $d_{(001)}$, the spacing between the planes. That, of course is the **Volume** of the unit cell. If we divide this quantity into the area, we get **the reciprocal of the spacing**, which is what we want!!

$$1/d_{(001)} = Area/Volume = |\mathbf{s}_{001}| = \left|{}^{\mathbf{a}\ \ \mathbf{b}}/_{\mathbf{a}\times\mathbf{b}\cdot\mathbf{c}}\right| = c^{*}$$

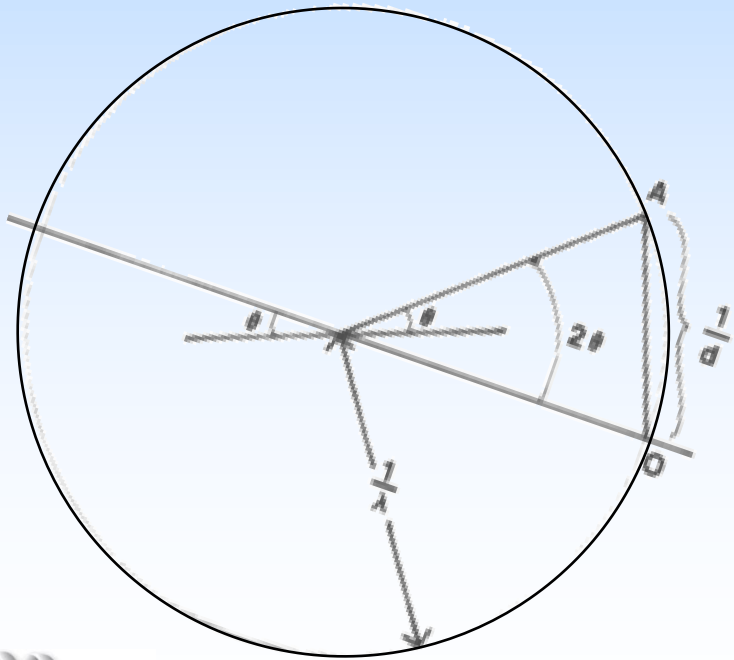


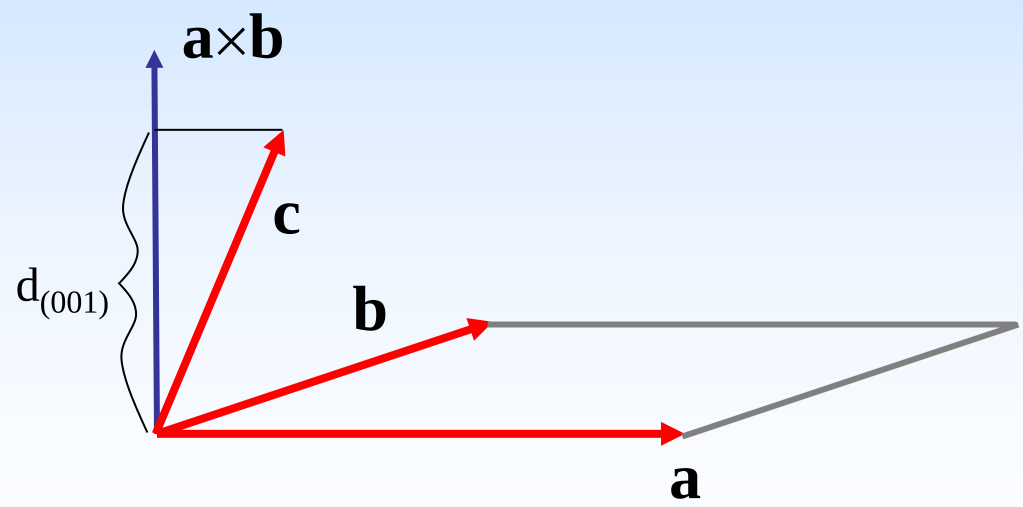
So the *reciprocal* lattice vector that represents the (001) planes is

$$\mathbf{s}_{001} = \mathbf{a}\times\mathbf{b} / \mathbf{a}\times\mathbf{b}\cdot\mathbf{c} \quad \text{and} \quad |\mathbf{s}_{001}| = 1/d_{(001)}$$

We define each axial reciprocal lattice vector as a reciprocal unit cell axis:

$$\mathbf{s}_{100} = \mathbf{a}^* \qquad \mathbf{s}_{010} = \mathbf{b}^* \qquad \mathbf{s}_{001} = \mathbf{c}^*$$





Let's be sure this is perfectly clear:

We define each **principal** reciprocal lattice vector as a **reciprocal unit cell axis**:

$\mathbf{a}^* = \mathbf{s}_{100} = \mathbf{b}\times\mathbf{c} / \mathbf{a}\times\mathbf{b}\cdot\mathbf{c}$ and $|\mathbf{s}_{100}| = 1/d_{(100)}$

$\mathbf{b}^* = \mathbf{s}_{010} = \mathbf{c}\times\mathbf{a} / \mathbf{a}\times\mathbf{b}\cdot\mathbf{c}$ and $|\mathbf{s}_{010}| = 1/d_{(010)}$

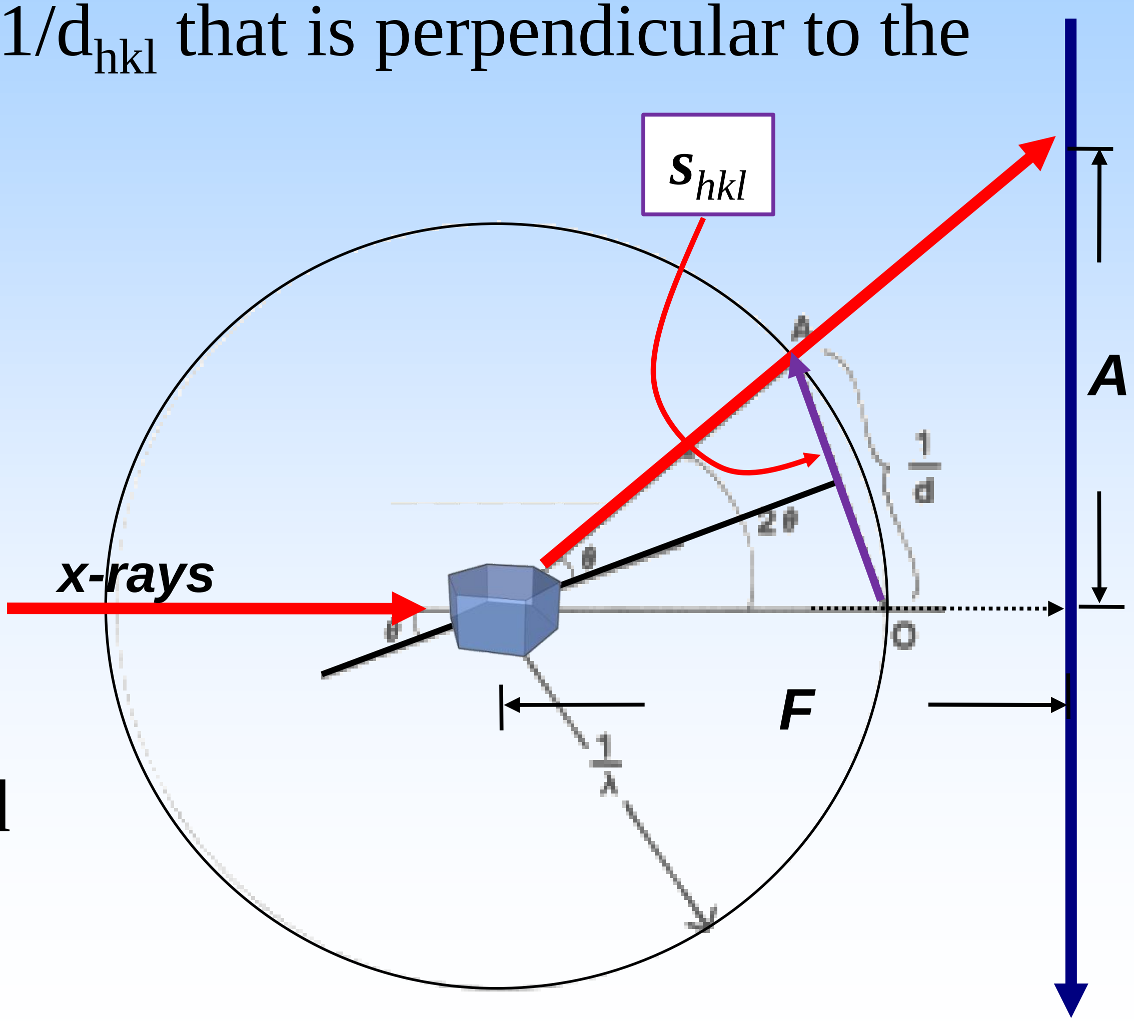
$\mathbf{c}^* = \mathbf{s}_{001} = \mathbf{a}\times\mathbf{b} / \mathbf{a}\times\mathbf{b}\cdot\mathbf{c}$ and $|\mathbf{s}_{001}| = 1/d_{(001)}$

These allow us to define reciprocal-lattice vectors:

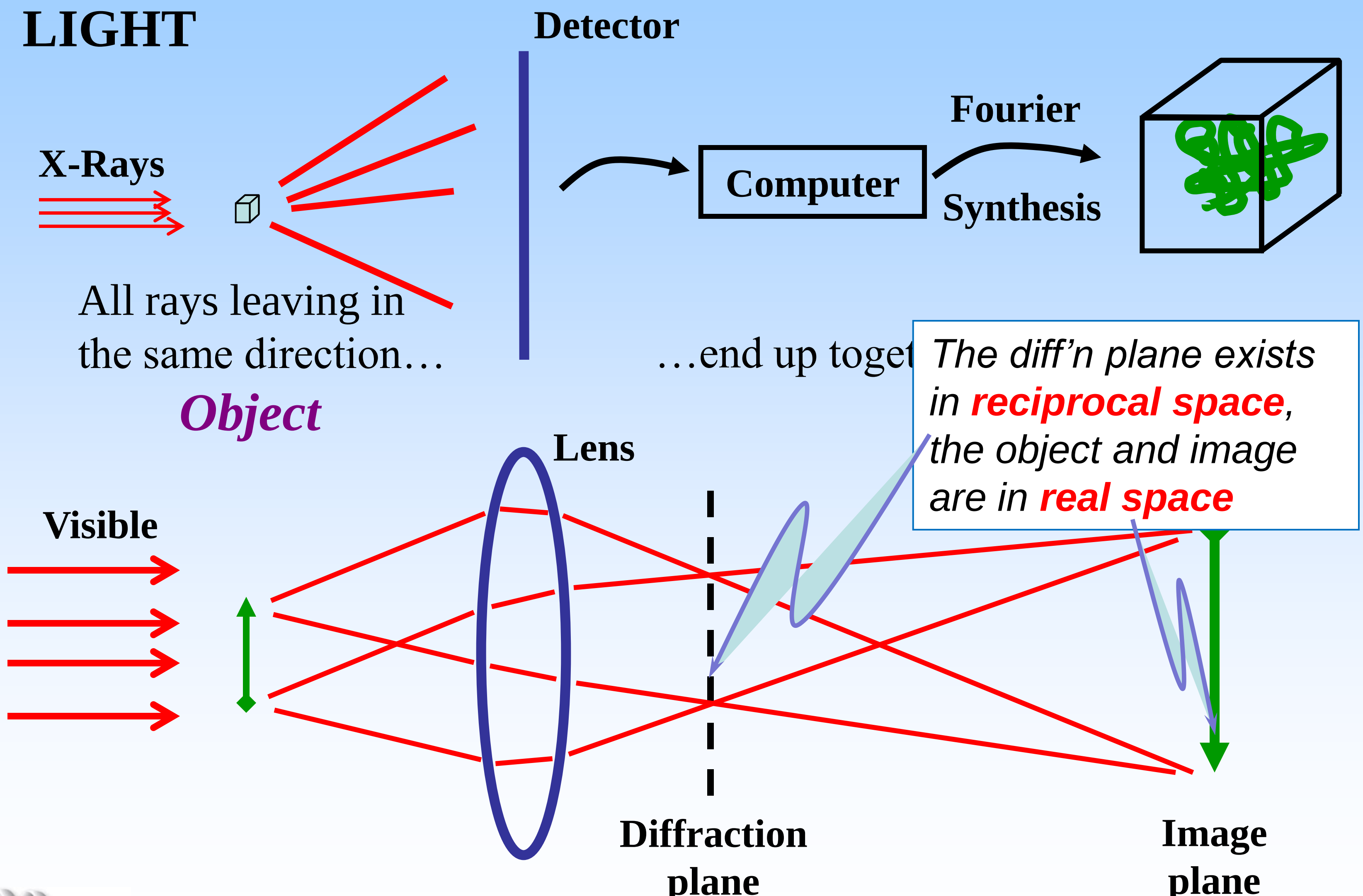
$$\mathbf{s}_{hkl} = h\mathbf{a}^* + k\mathbf{b}^* + l\mathbf{c}^*$$

Bragg's Law is obeyed – diffraction will occur – when the **s** vector of length $1/d_{hkl}$ that is perpendicular to the lattice plane (hkl) touches the Ewald sphere of radius $1/\lambda$.

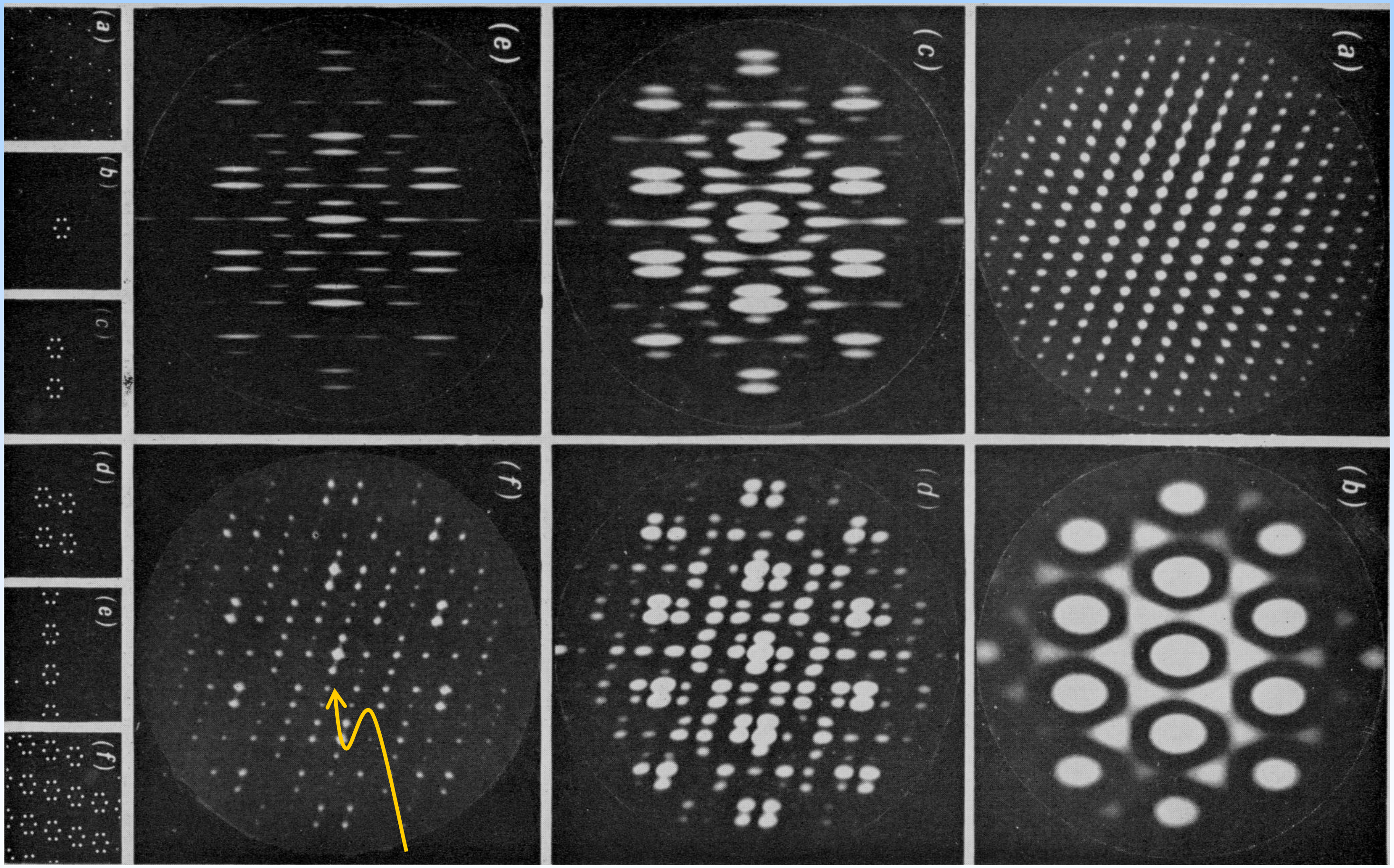
So we need not think about Bragg planes again, we think only of reciprocal-lattice vectors and the Ewald Sphere of reflection.



Remember our comparison between diffraction and lens imaging…



Now we use the Taylor and Lipson figures to see how the **contents** of the crystal relate to the diffraction pattern.



crystal

Coordinates of points are “indices.”

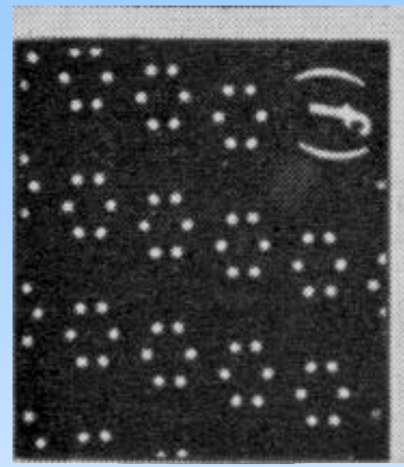
diffraction

Notice (1) The symmetry, and (2) how the continuous diffraction pattern of one molecule (b) is “sampled” by the lattice of diffraction points.

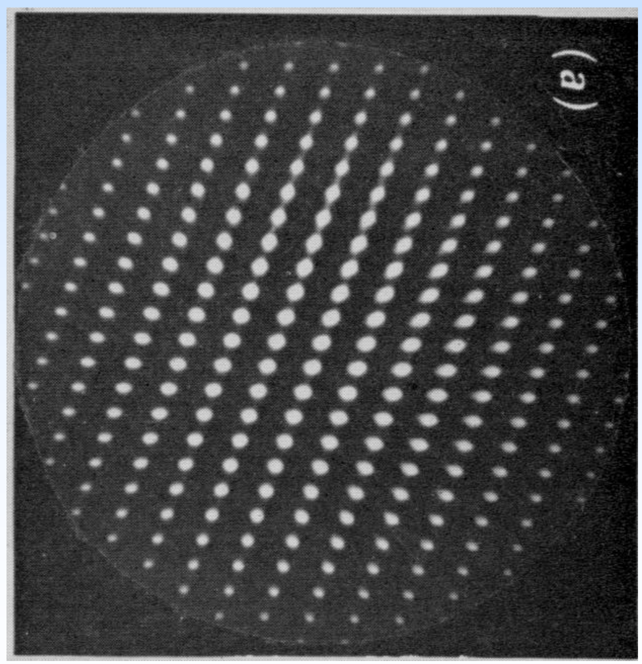
Plate 2 from Taylor and Lipson -- Optical Transforms

Do we understand the real/reciprocal lattice idea?

Crystal –
Real Lattice



Diffraction –
Reciprocal Lattice



Confirm that the vectors perpendicular to the **Crystal-Lattice planes** are parallel to the **Reciprocal Lattice vectors**, and that the reciprocal distances make sense.

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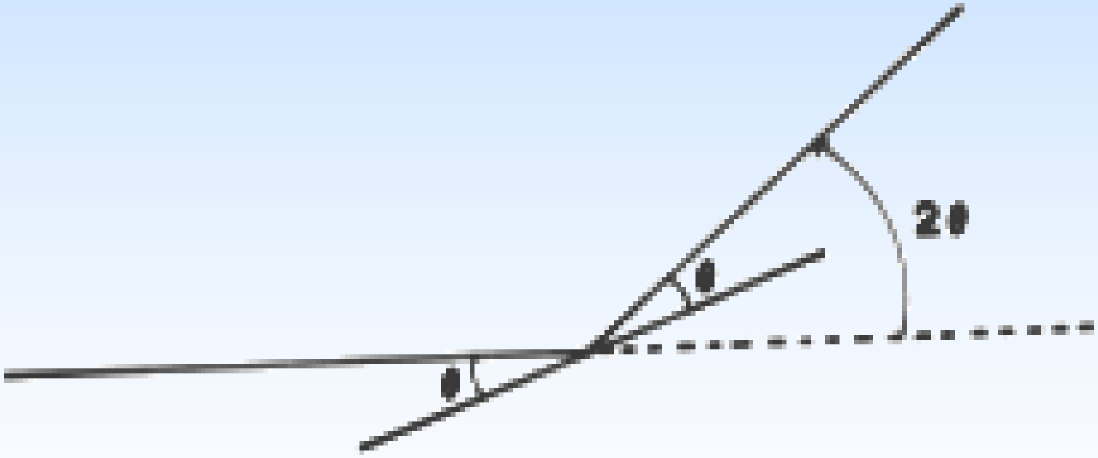
- Outline the structure-solving methods -- heavy atoms and MADness

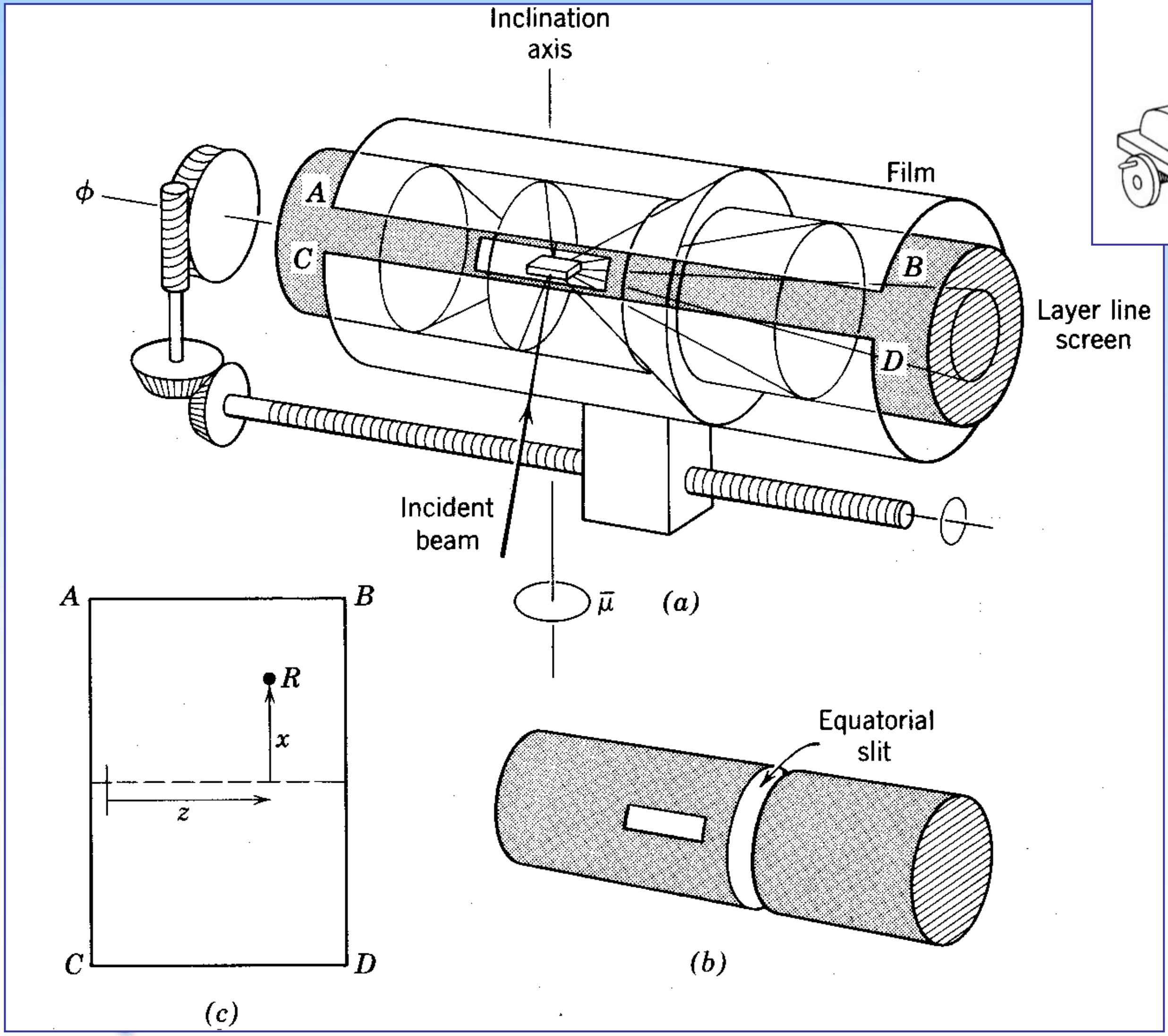
Remember the geometry -- if the **Bragg** planes lie angle θ from the incident x-ray beam, the **total diffraction angle will be 2θ**. We can make an instrument to exploit that geometry.

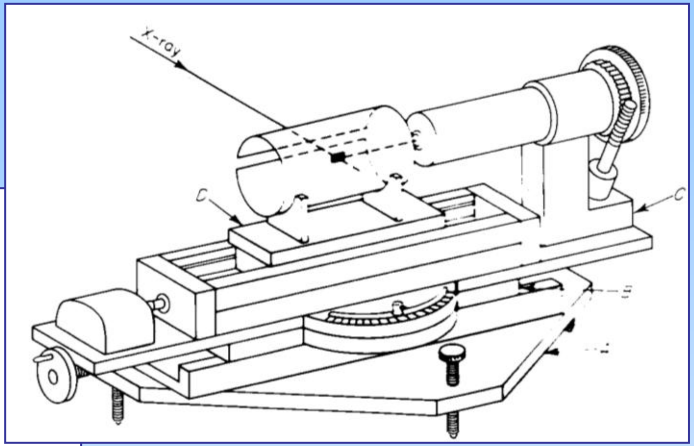


And Remember the objective – We must view the molecule from every direction to recreate a three-dimensional image:

- We must obtain diffraction from *all* of the **Bragg** planes;
- We must sample *all* of the **reciprocal lattice**.

My first data were collected with a Weissenberg Camera *47-yrs ago.*





A complicated machine to simplify our view of reciprocal space.

One rotates the crystal around a ***real lattice vector.***

The Weissenberg photograph gives a wonderfully distorted, but organized, view of reciprocal space.

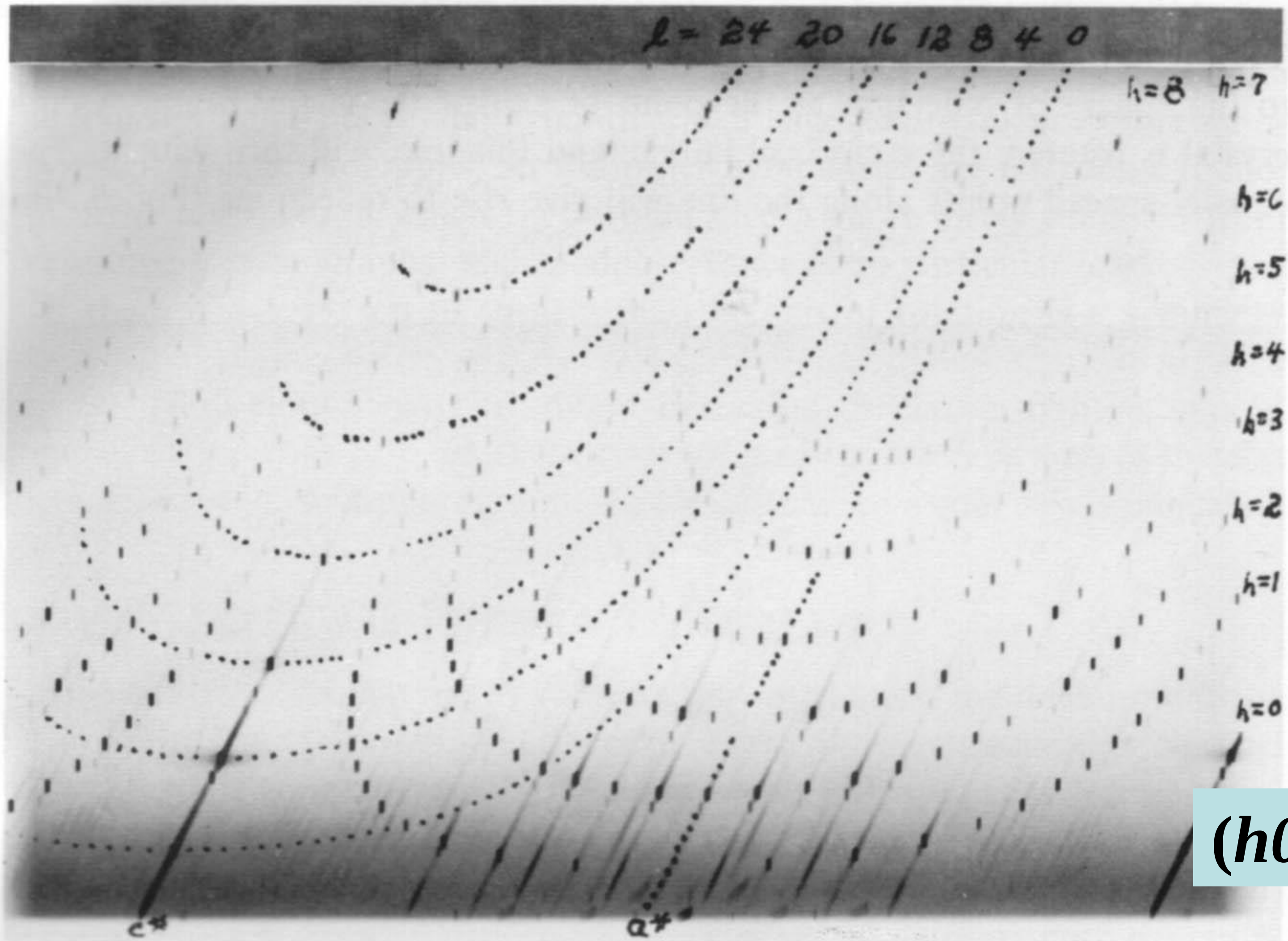
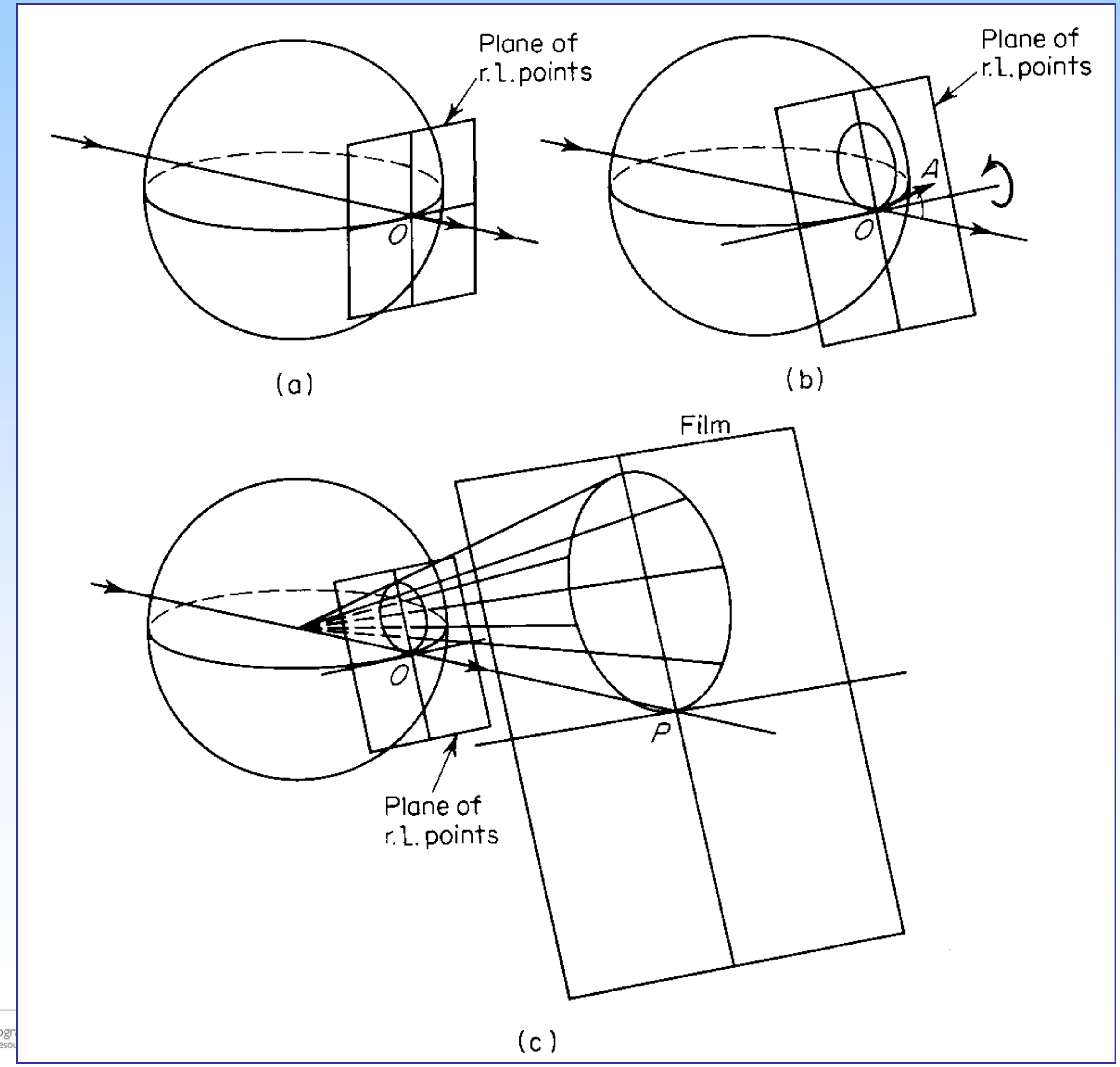


Figure 5.23. Weissenberg photograph showing indexed reciprocal lattice lines.

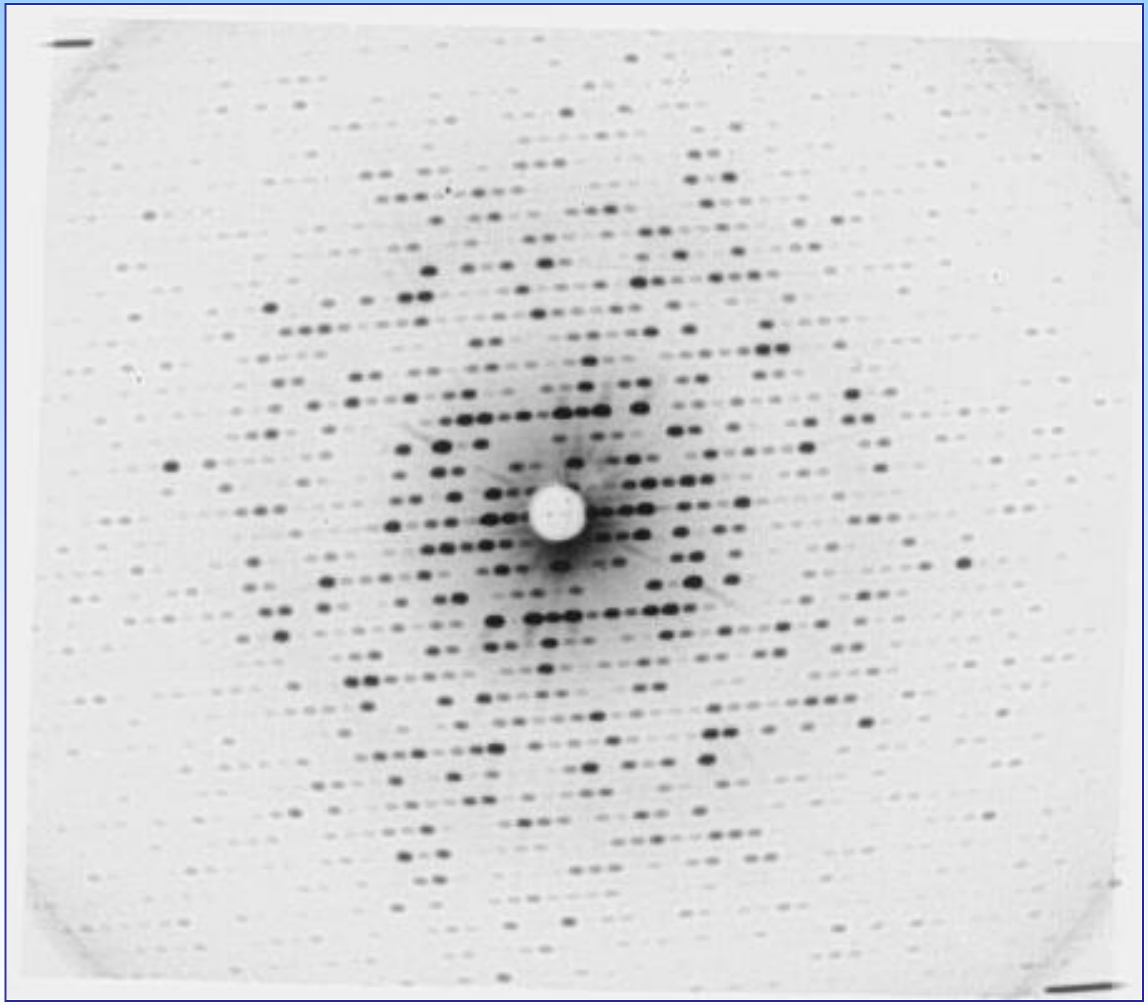
(h0l) data

Martin Buerger devised a camera geometry that preserved the shape of reciprocal space.



It's an even more complicated machine to simplify our view of reciprocal space further.

Mount x-tals with a ***real*** *crystal axis* ***parallel to the beam.***



An antique precession photo of Chymotripsin, courtesy of David M Blow

The **precession photograph** allows us to view the diffraction pattern of the crystal lattice as an **undistorted** pattern of spots.

How can we lear

cell from a prece

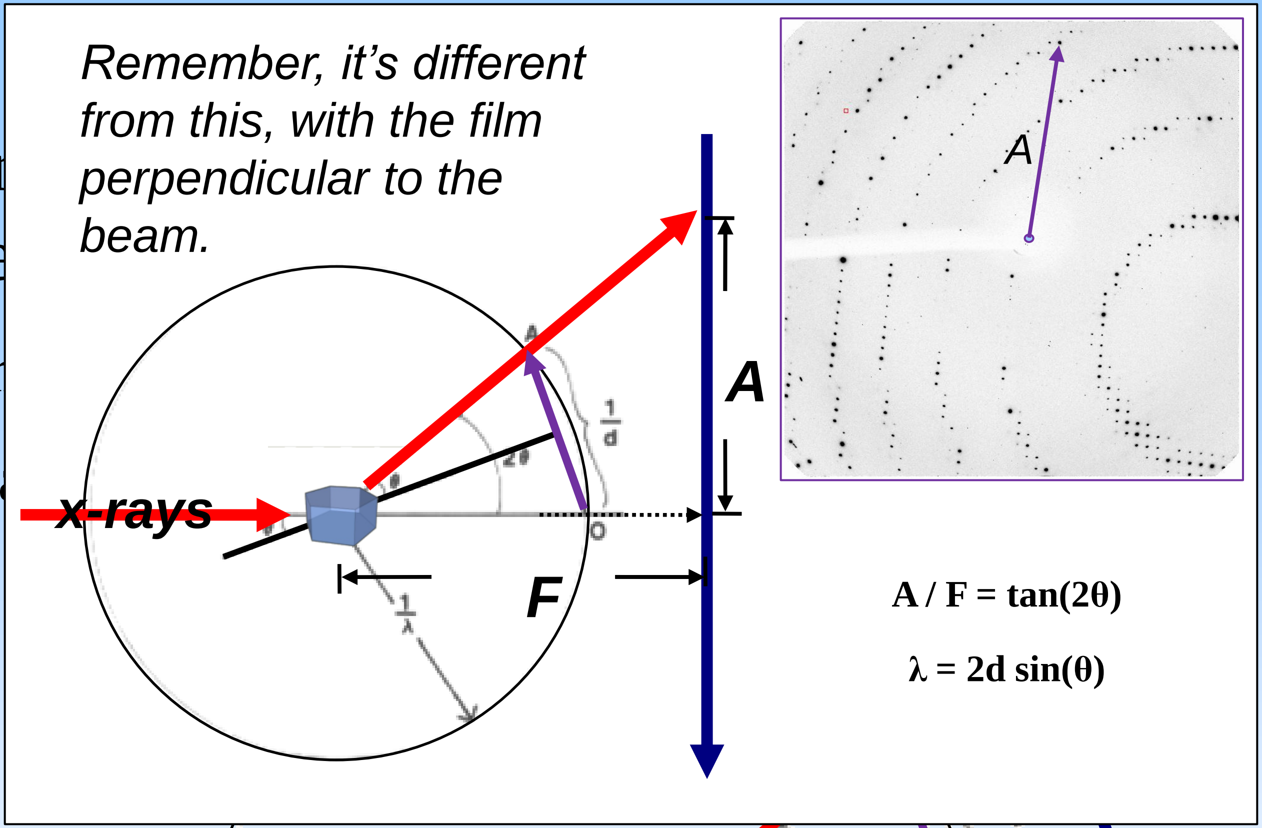
It’s easy, because th

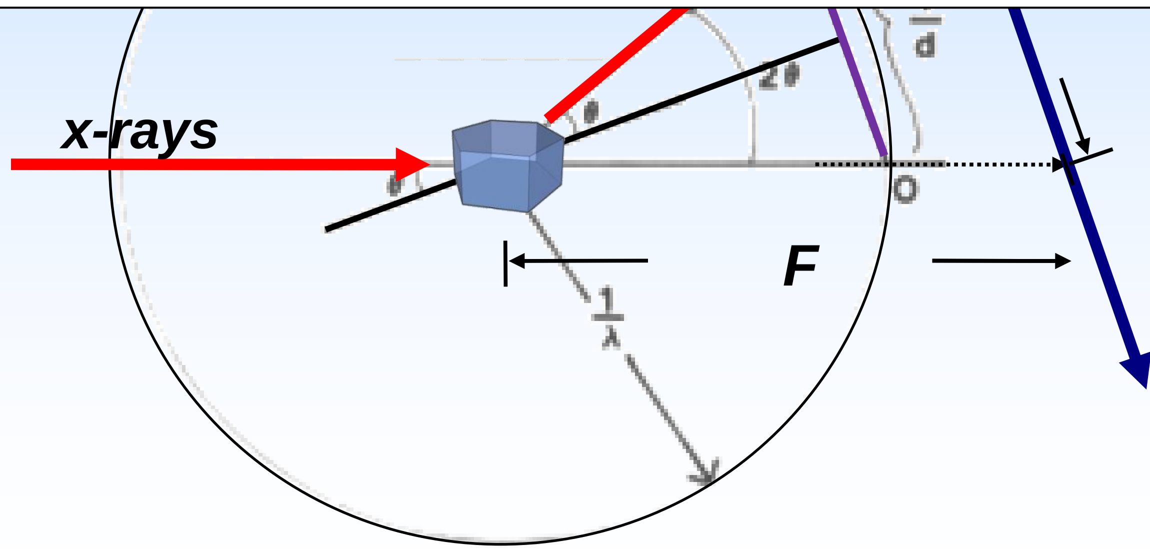
So from this we ha

$$B/F = {}^{(1/d)}/_{(1/\lambda)}$$

Or

$$d_{(hkl)} = \lambda F/B$$





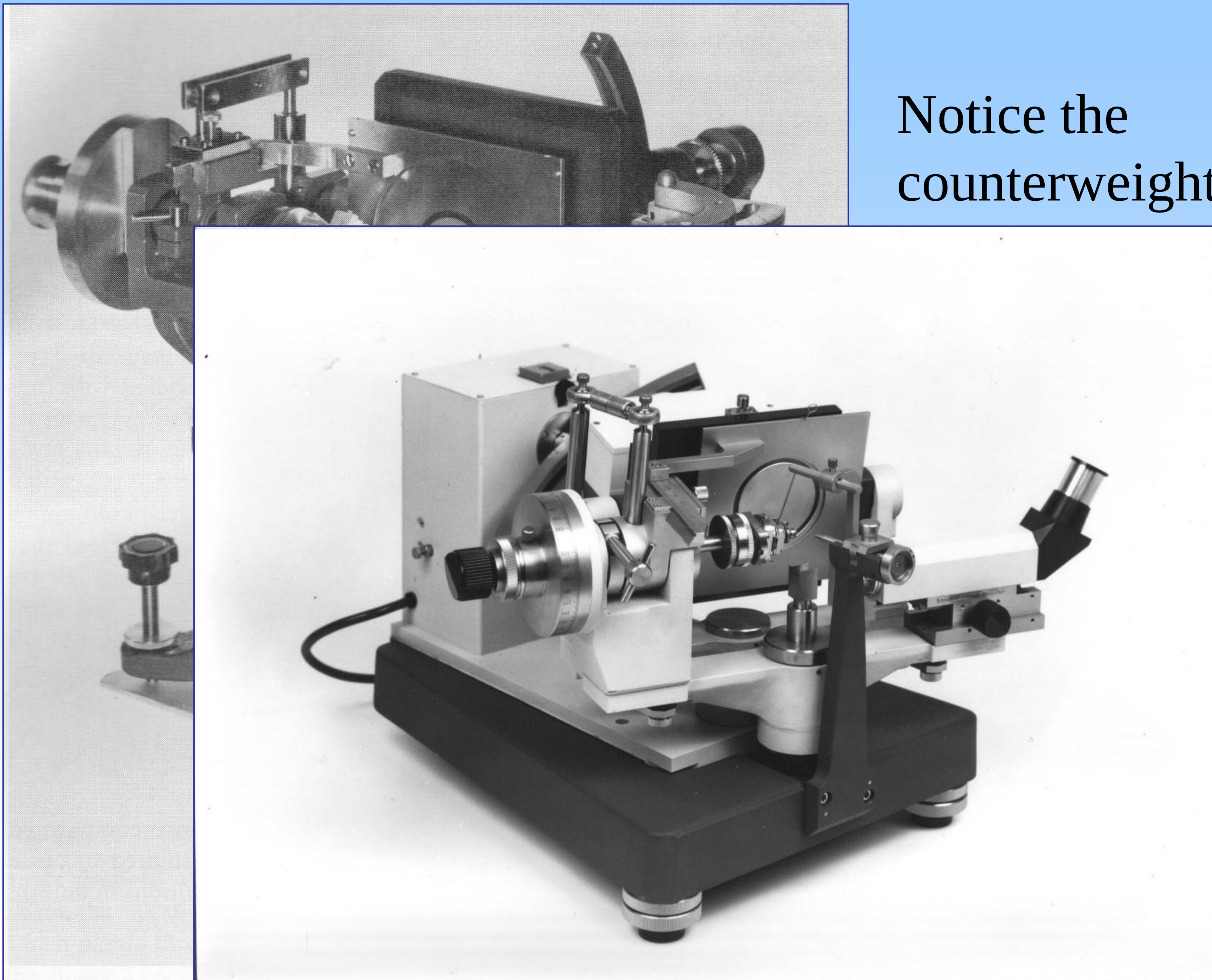


Figure 5.59. Precession camera. (Co

Bram Schierbeek, Bruker-AXS

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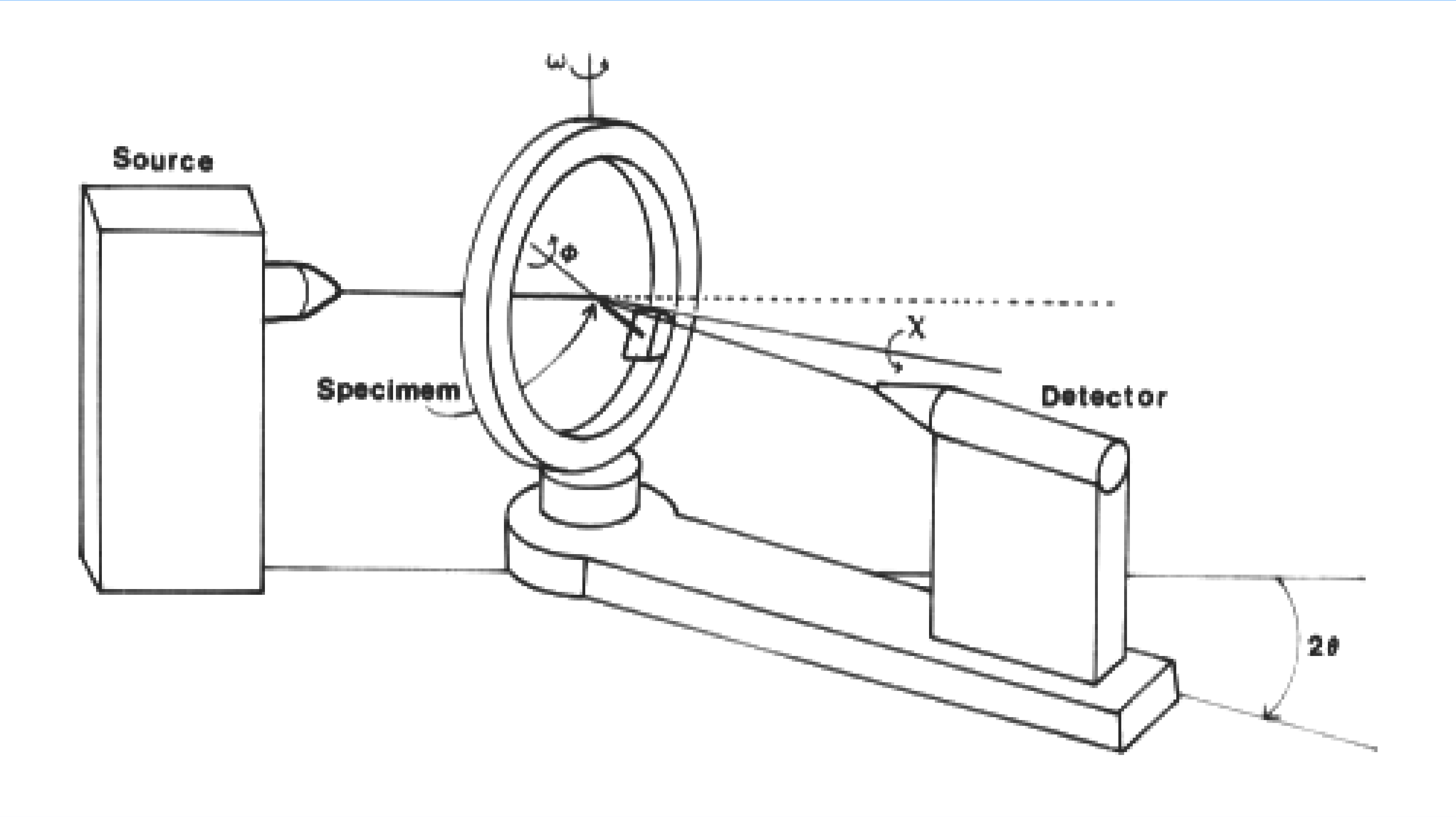
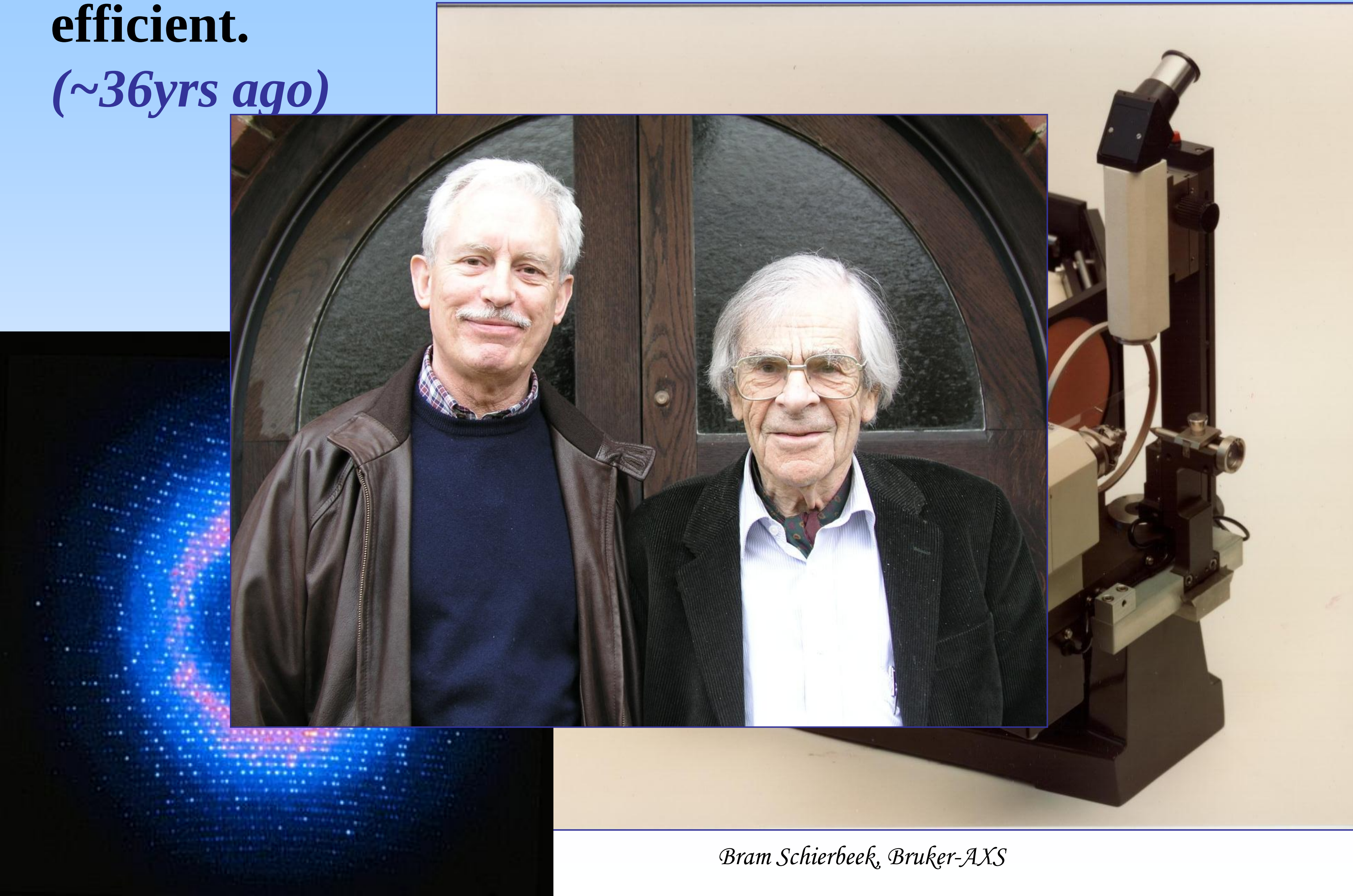
The automated Eulerian cradle decreased the labor, but still one measured reflections one at a time. *(45yrs ago)*



Uli Arndt and Alan Wonacott invented the automated rotation camera. Still x-ray film, but very much more efficient.

(~36yrs ago)



Bram Schierbeek, Bruker-AXS

Another Uli Arndt invention was a video-based detector

The screen was small, but it was very sensitive and could read out continuously – the x-tal just kept rotating as images came out.

(23yrs ago)

Bram Schierbeek
Bruker-AXS

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An important advance was photoluminescent imaging plates. MAR research, followed by Rigaku, made a successful camera that worked like electronic x-ray film, but much better. *(21 yrs)*

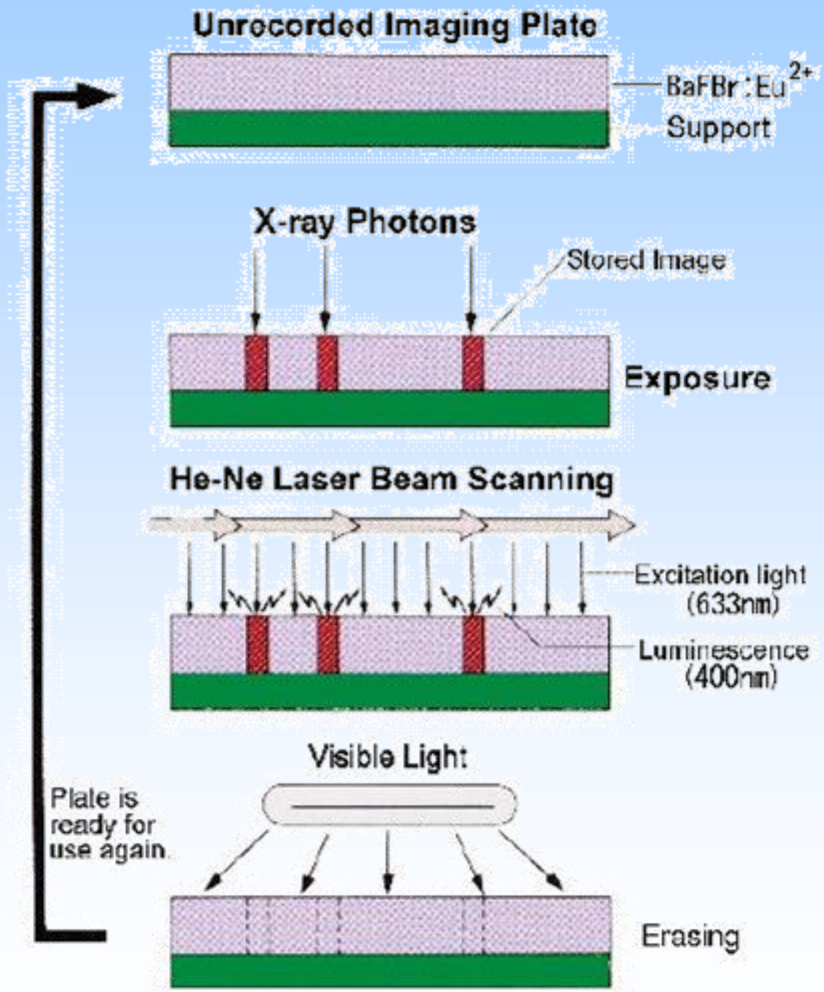
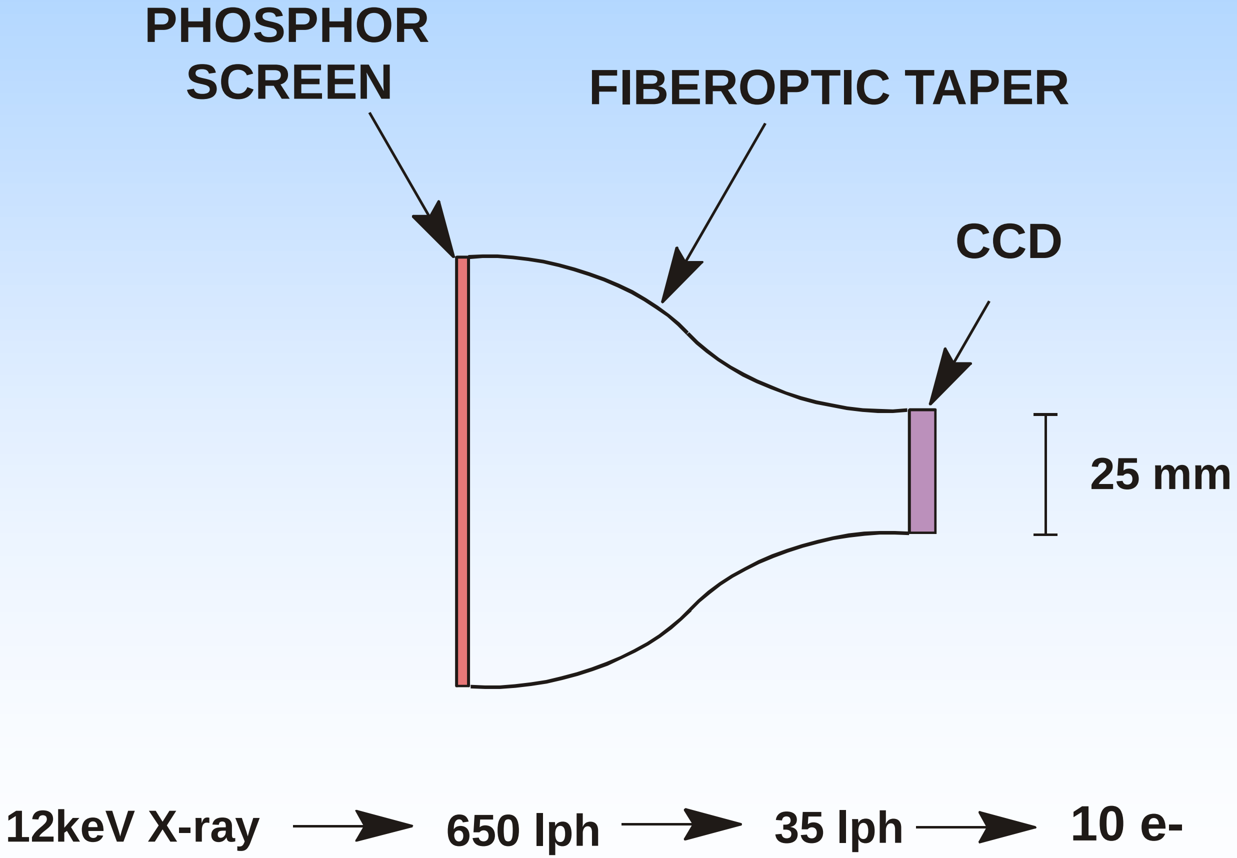
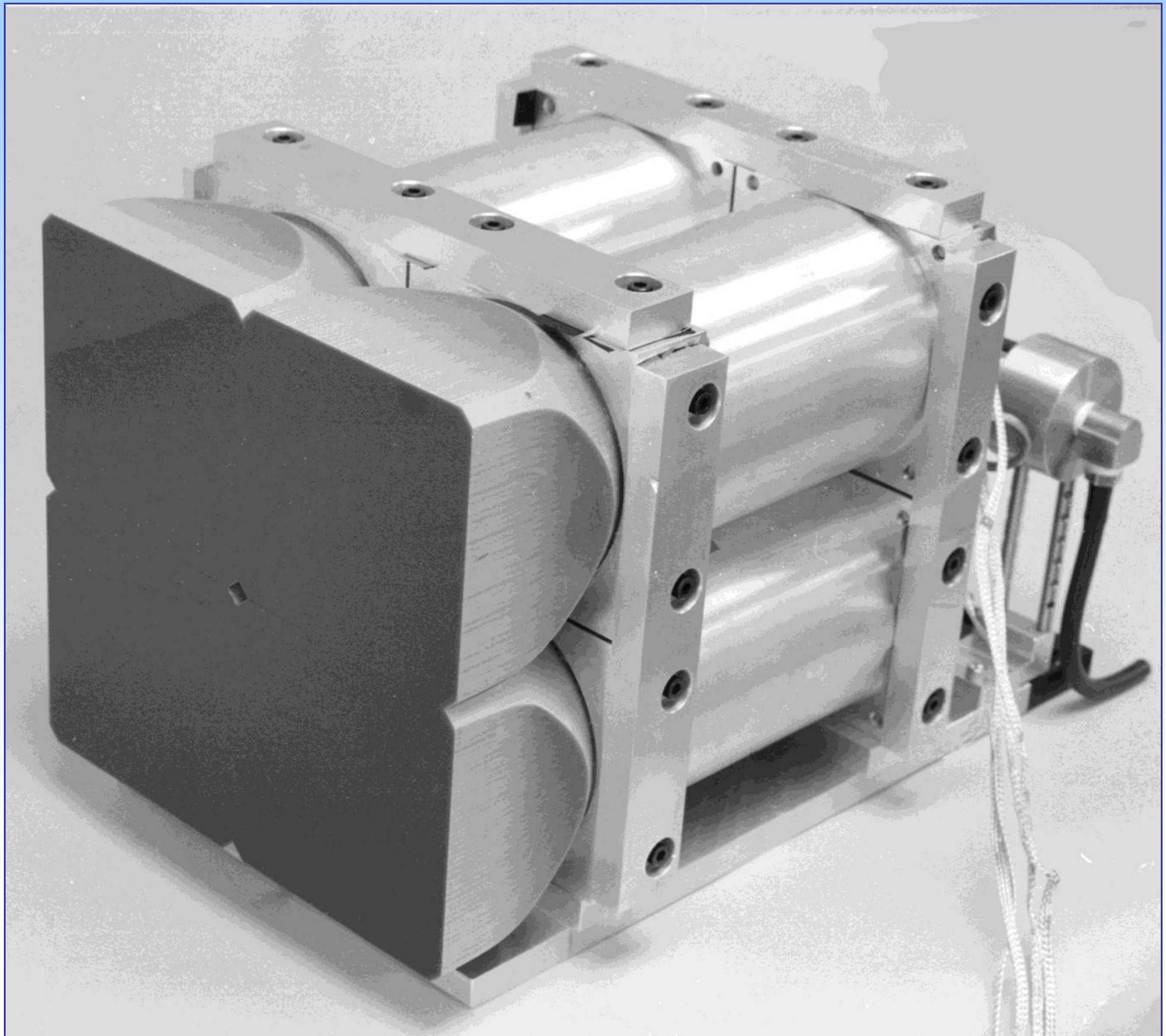


Figure 1. The process of recording an x-ray radiation image on an imaging plate, and subsequent read-out and erasure.

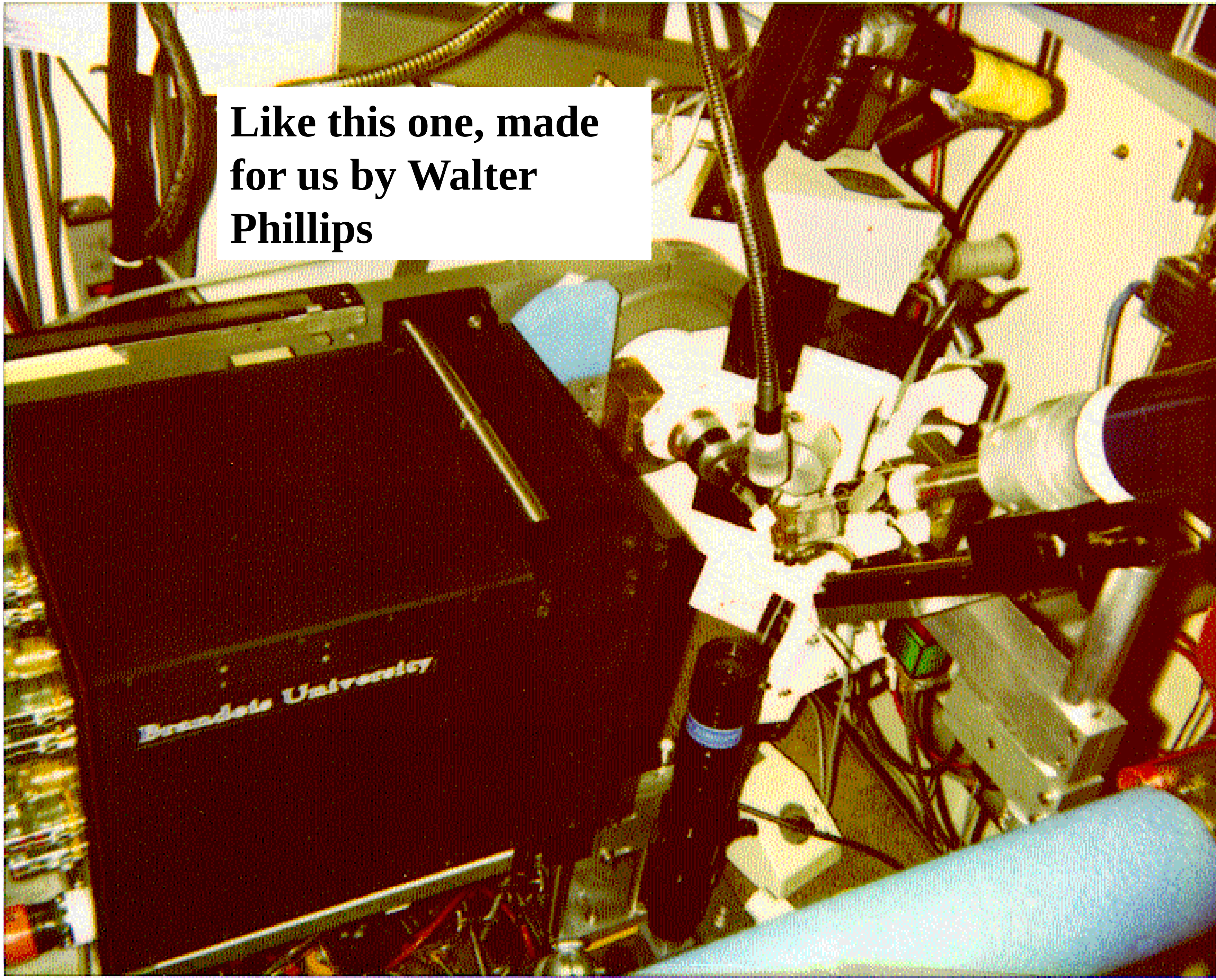
The advance that made possible our modern detectors was made by Ed Westbrook, Sol Gruner, and others: bonding of a charge-coupled device to a fiber-optic taper with an x-ray sensitive phosphor in front. ***(17yrs ago)***



Several of these can be bonded together to make a large detector… *(13yrs ago)*

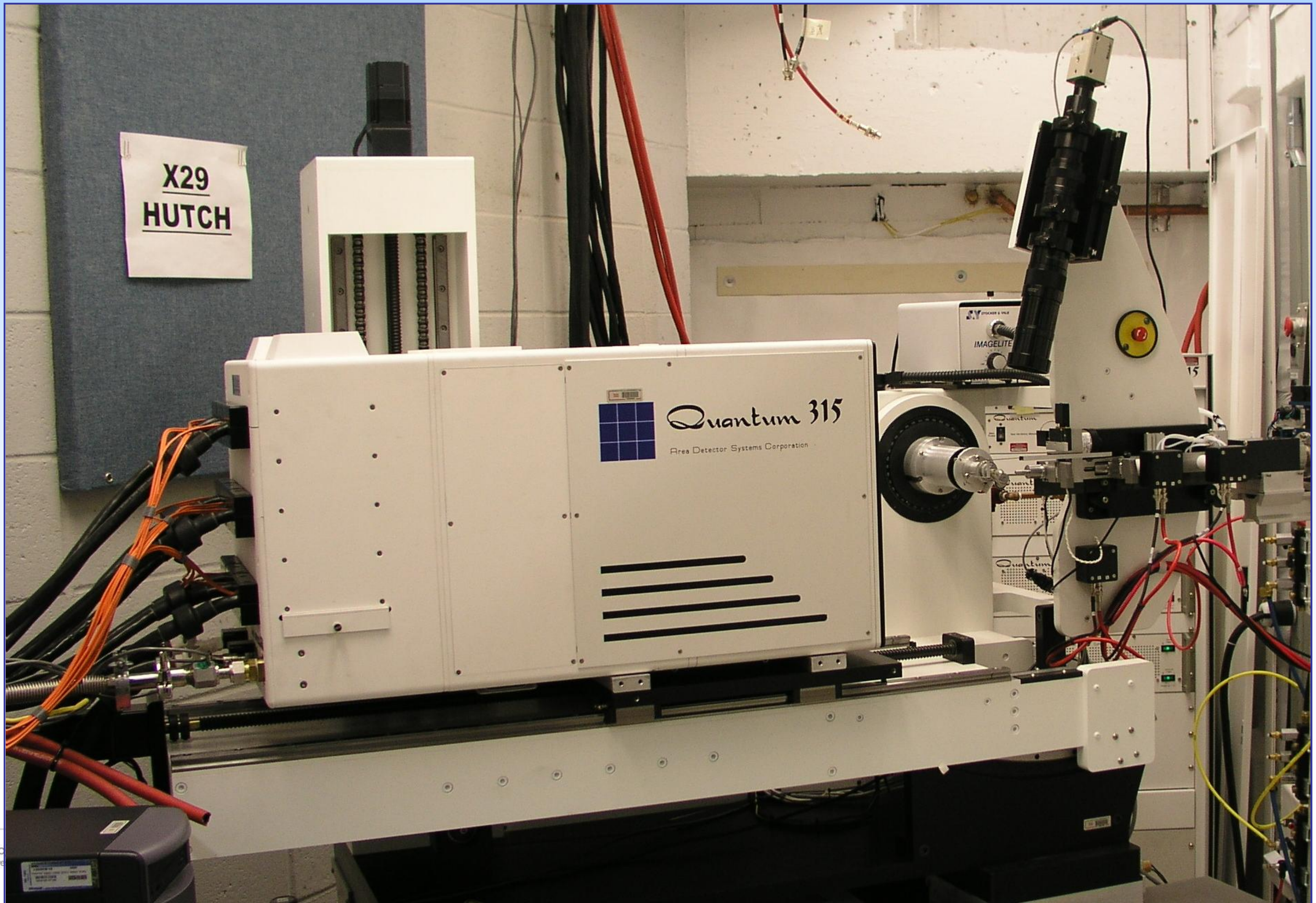


Like this one, made for us by Walter Phillips



And the modern commercial versions are large, fast, and very accurate. *(8yrs ago)*

Detectors like these are the basis for modern, high-throughput crystallography!



This is the next generation. It is noise free, and can produce images at 10 per second. *(1yr ago)*

This detector will be supplanted in 2-3 years by one that frames at 10,000 Hz!

The Pilatus 6M

(six million pixels)
Made by Dectris, a spin-off of the Paul Scherer Inst. (Switzerland's answer to Brookhaven National Laboratory)

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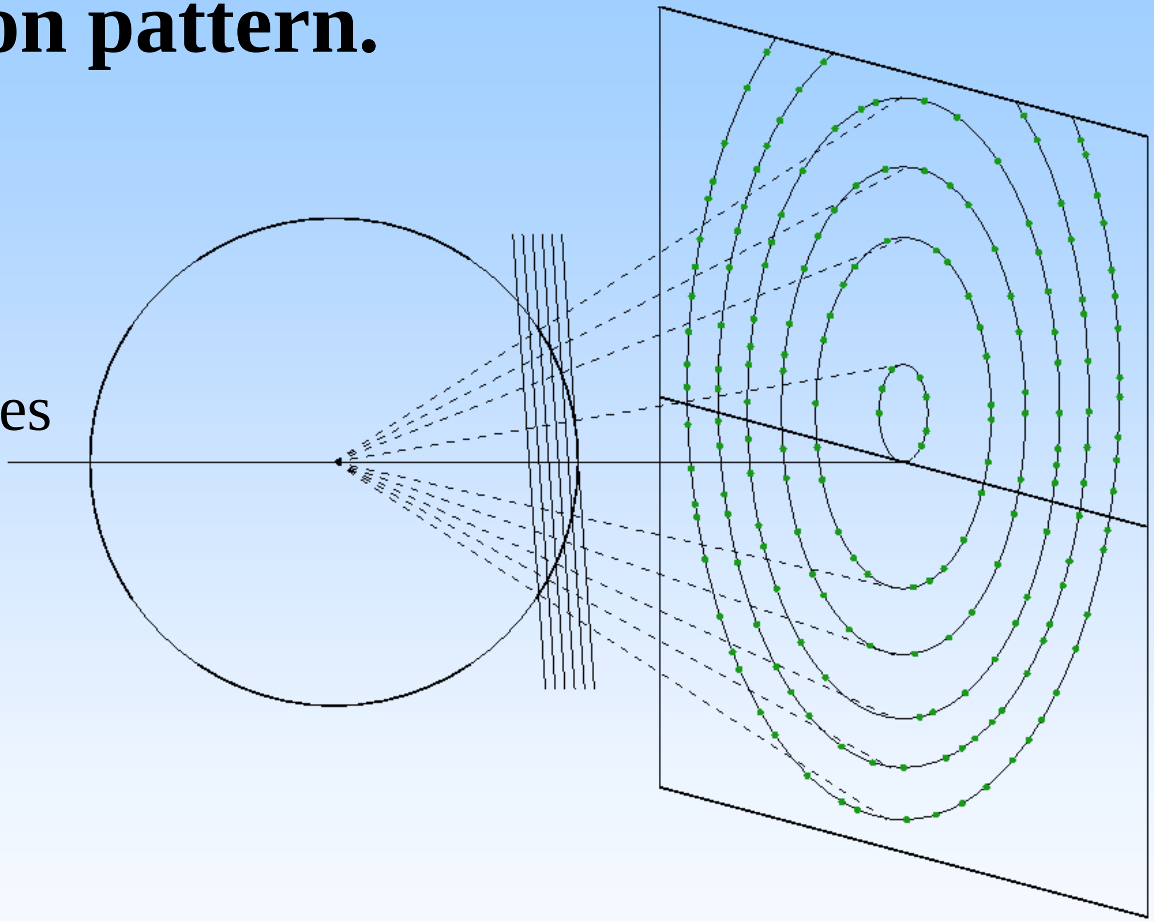
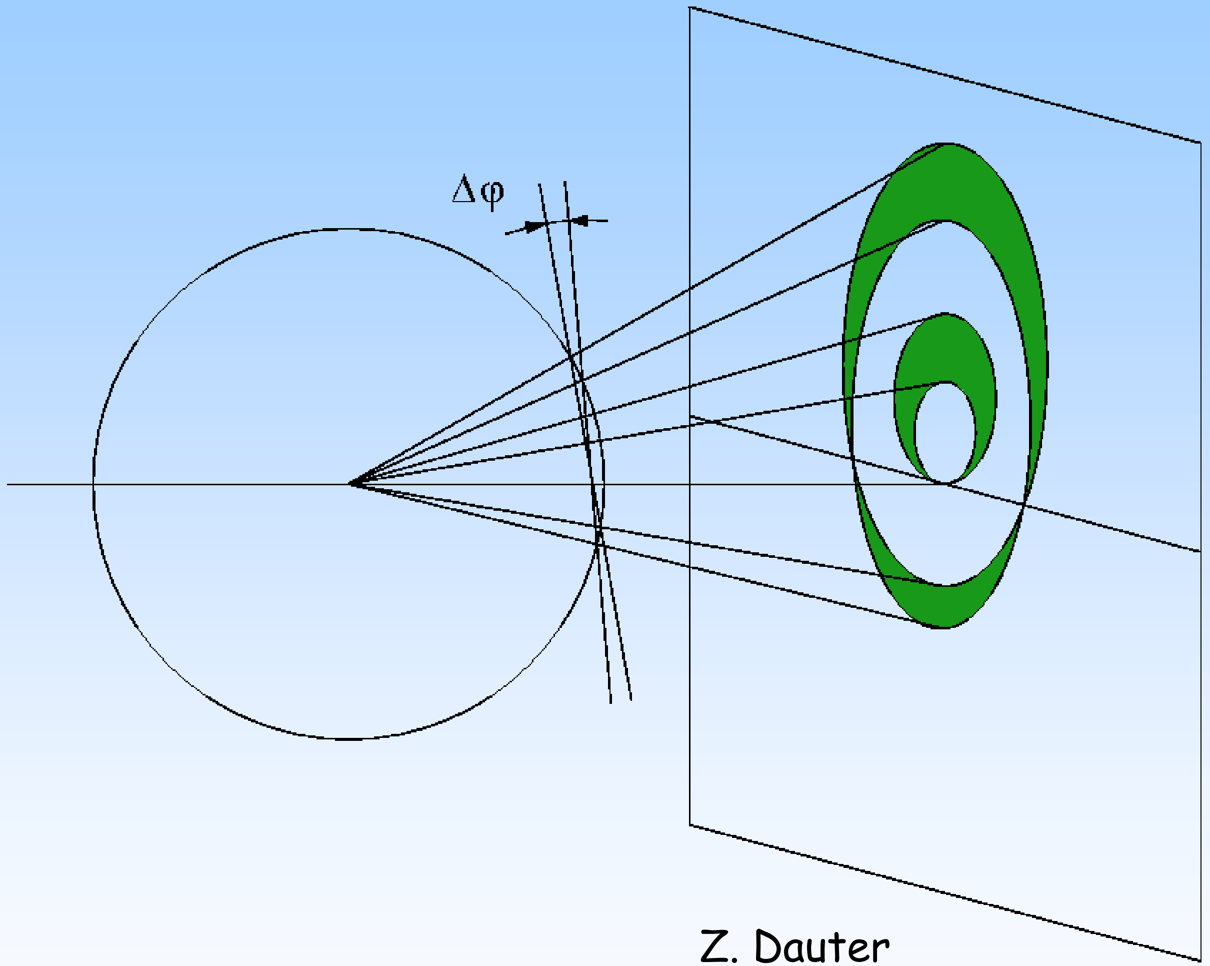
Look at how the reflections are generated in the diffraction pattern.

The planes of spots in reciprocal space appear as circles of spots on an area-sensitive x-ray detector (film, IP, CCD-based, *etc.*)



Z. Dauter

As the crystal is rotated, the circles are extended into “lunes”



Z. Dauter

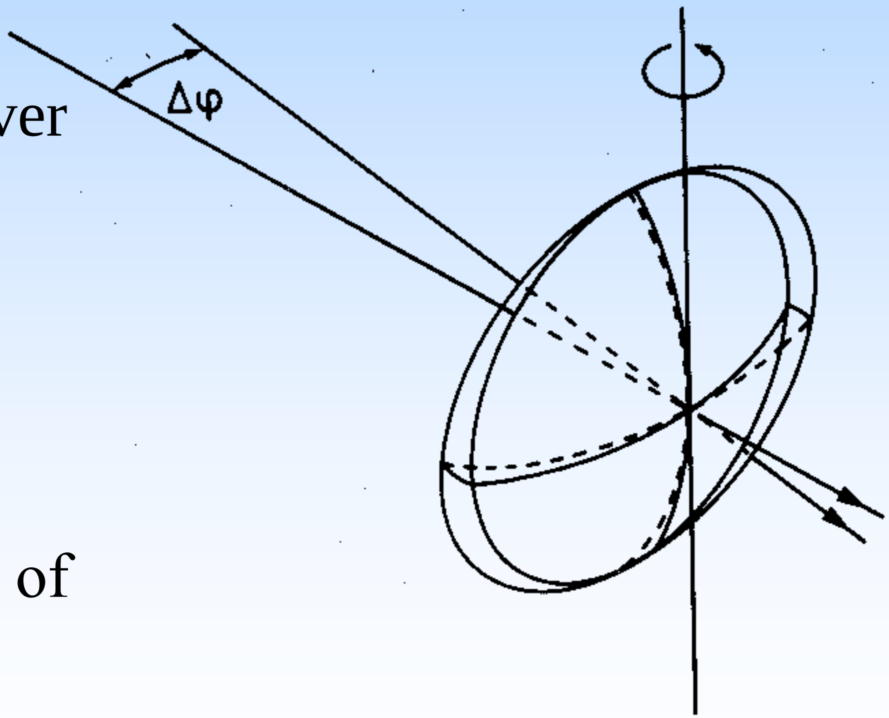
Rotation sweeps out a strangely-shaped volume. However...

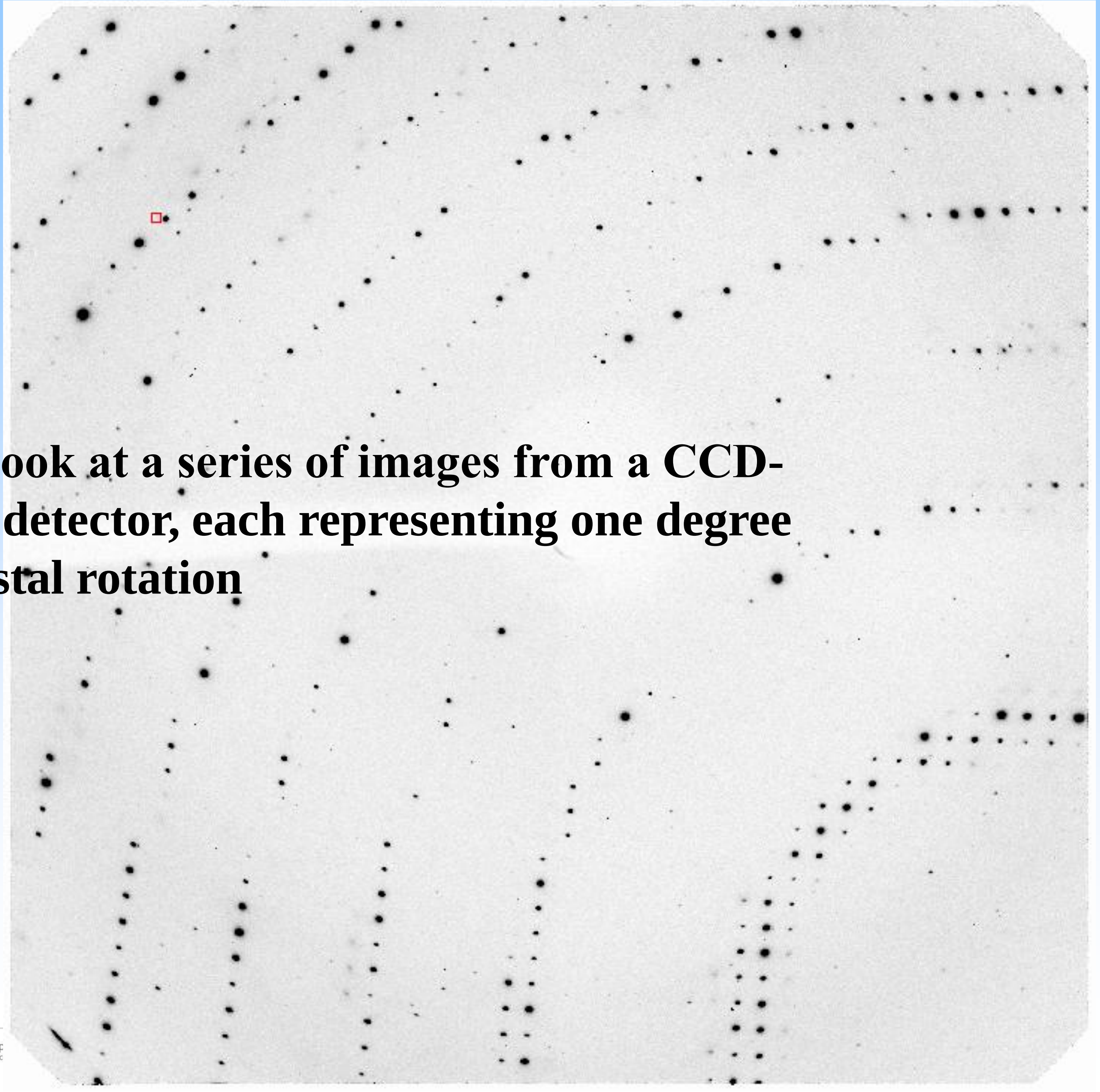
• Many r.l. points will be recorded during a single short rotation.

• Contiguous rotations will cover much of the reciprocal lattice.

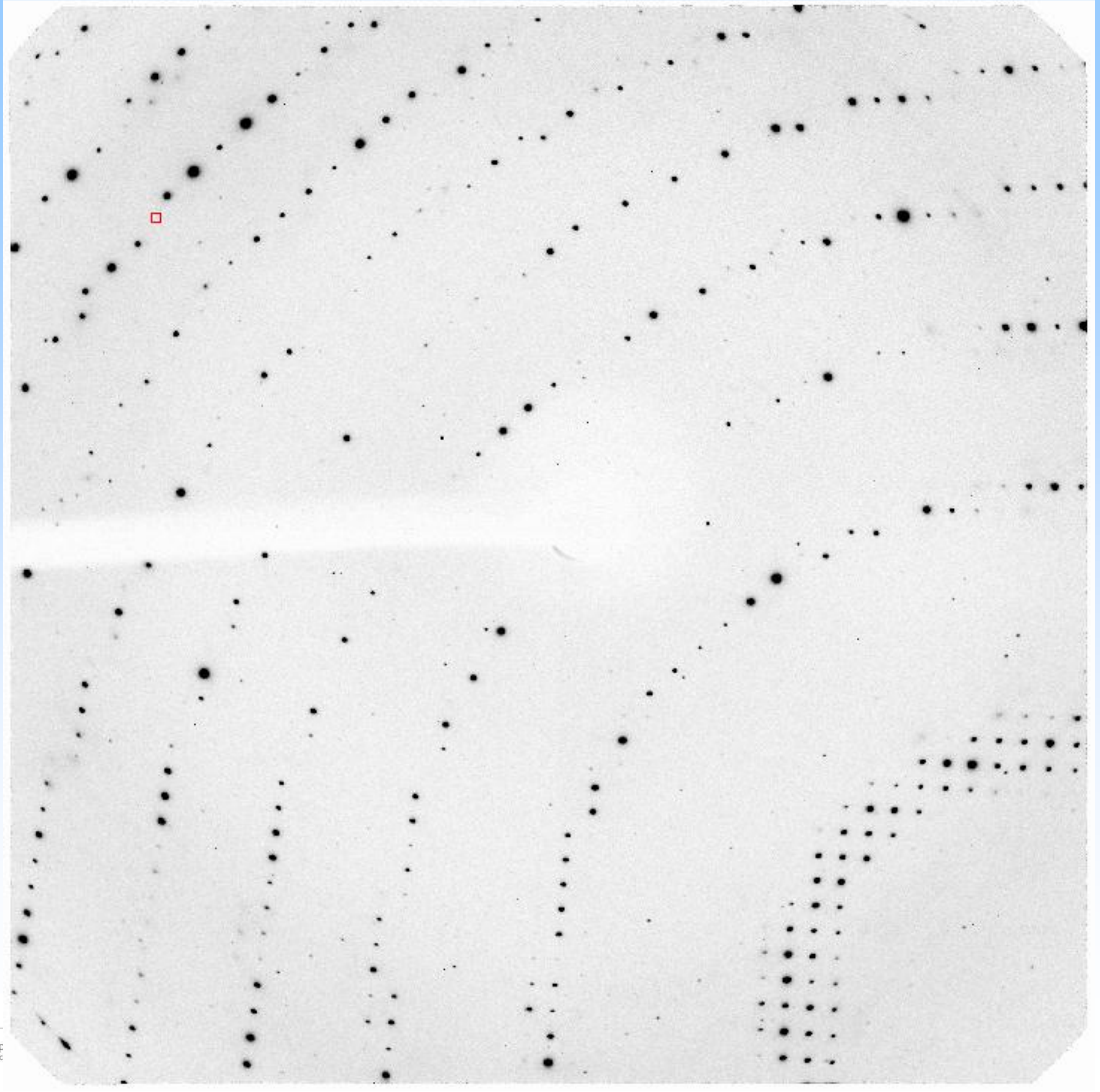
• The “camera” is simple: an axis, a film (or electronic detector), and a shutter.

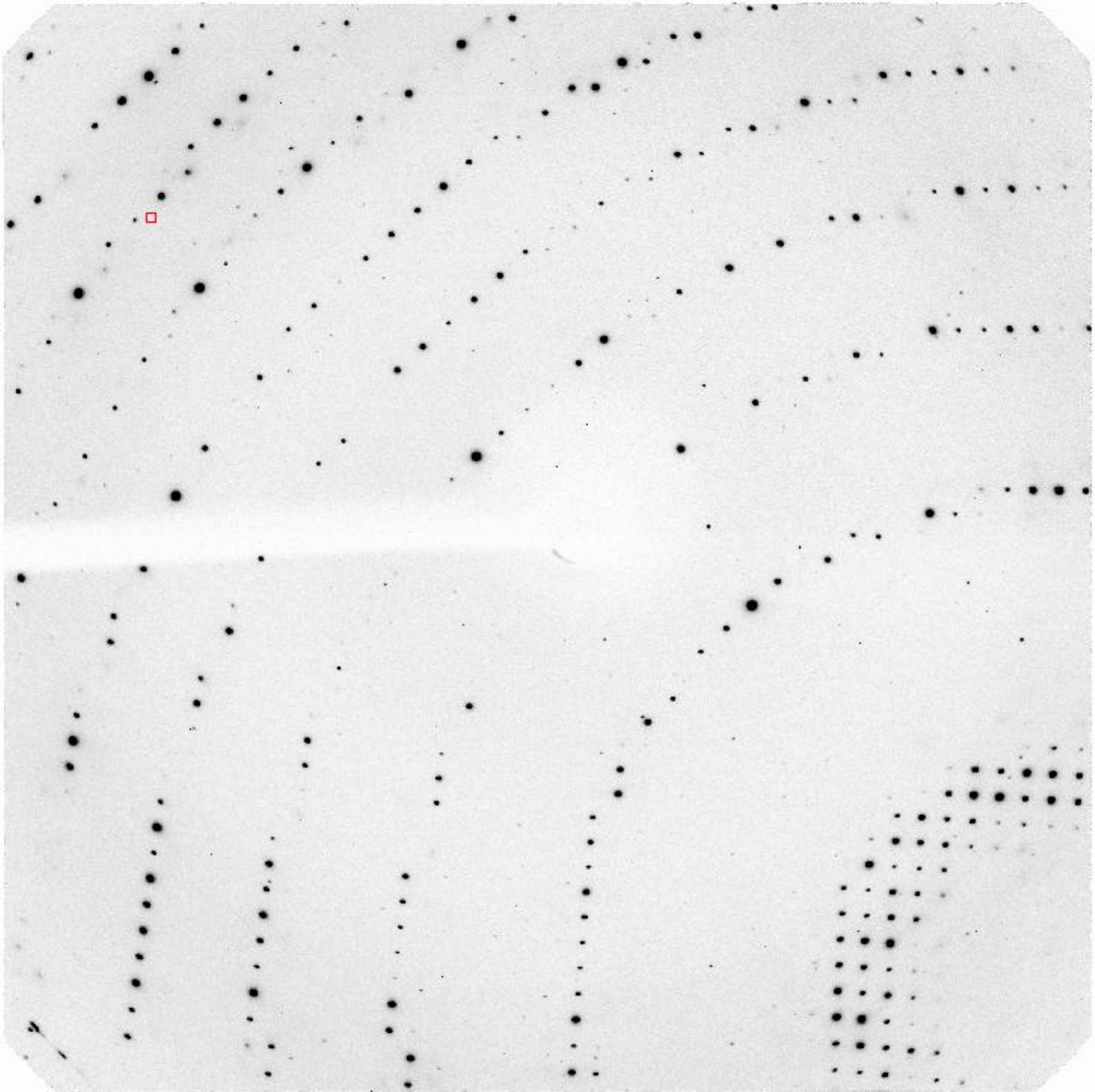
• It’s easy to substitute a range of detectors.





Let's look at a series of images from a CCD-based detector, each representing one degree of crystal rotation





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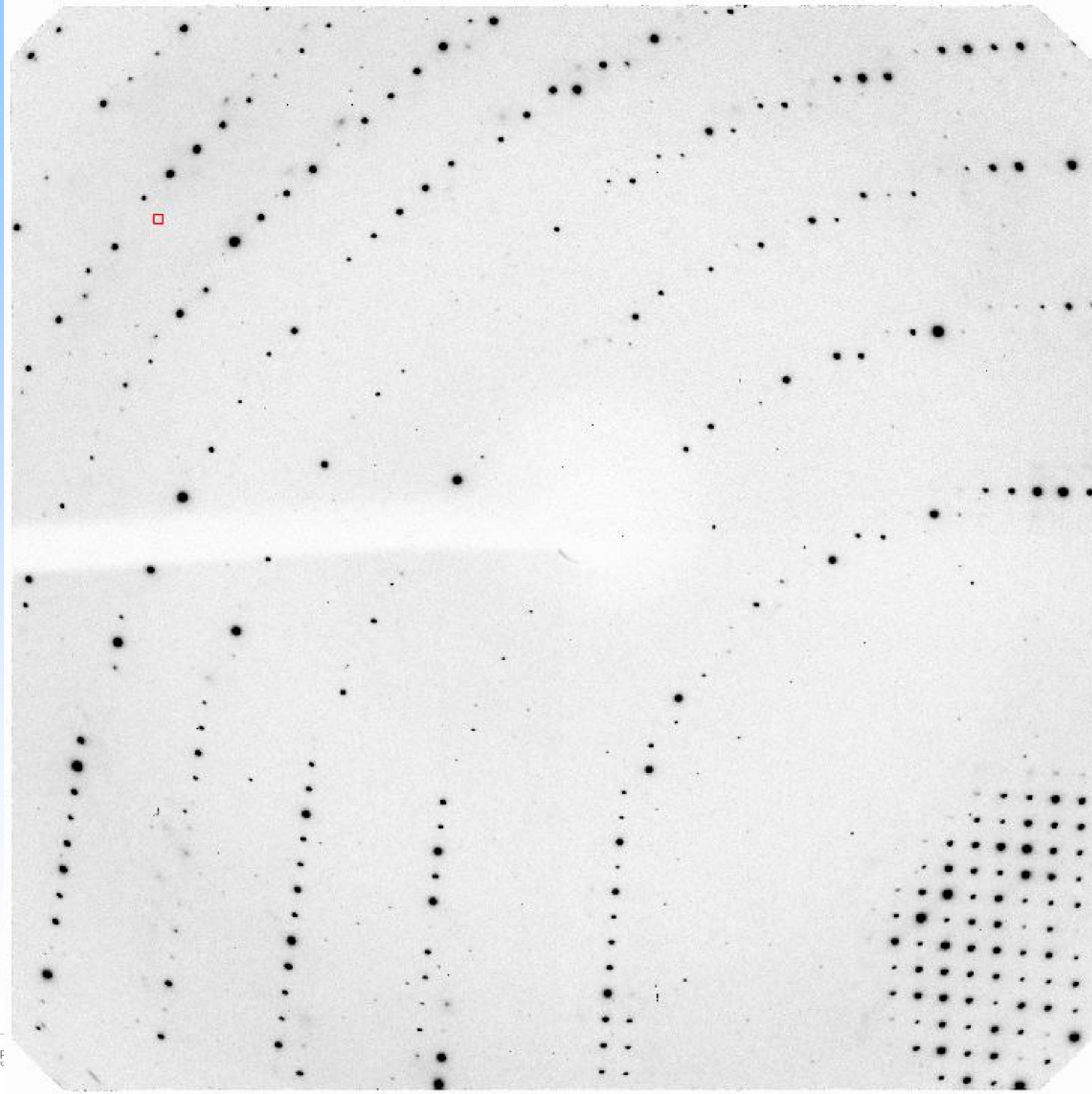

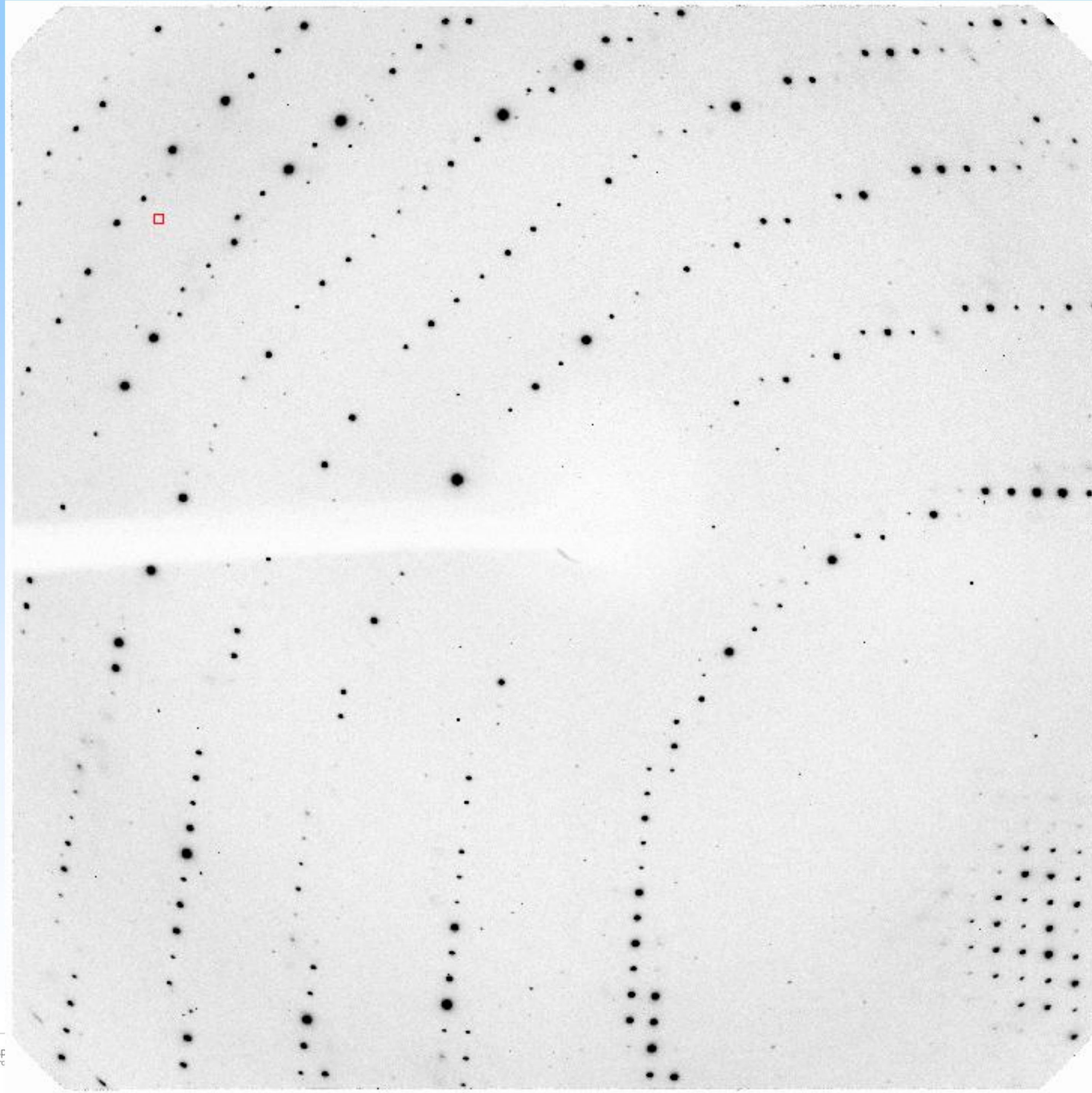

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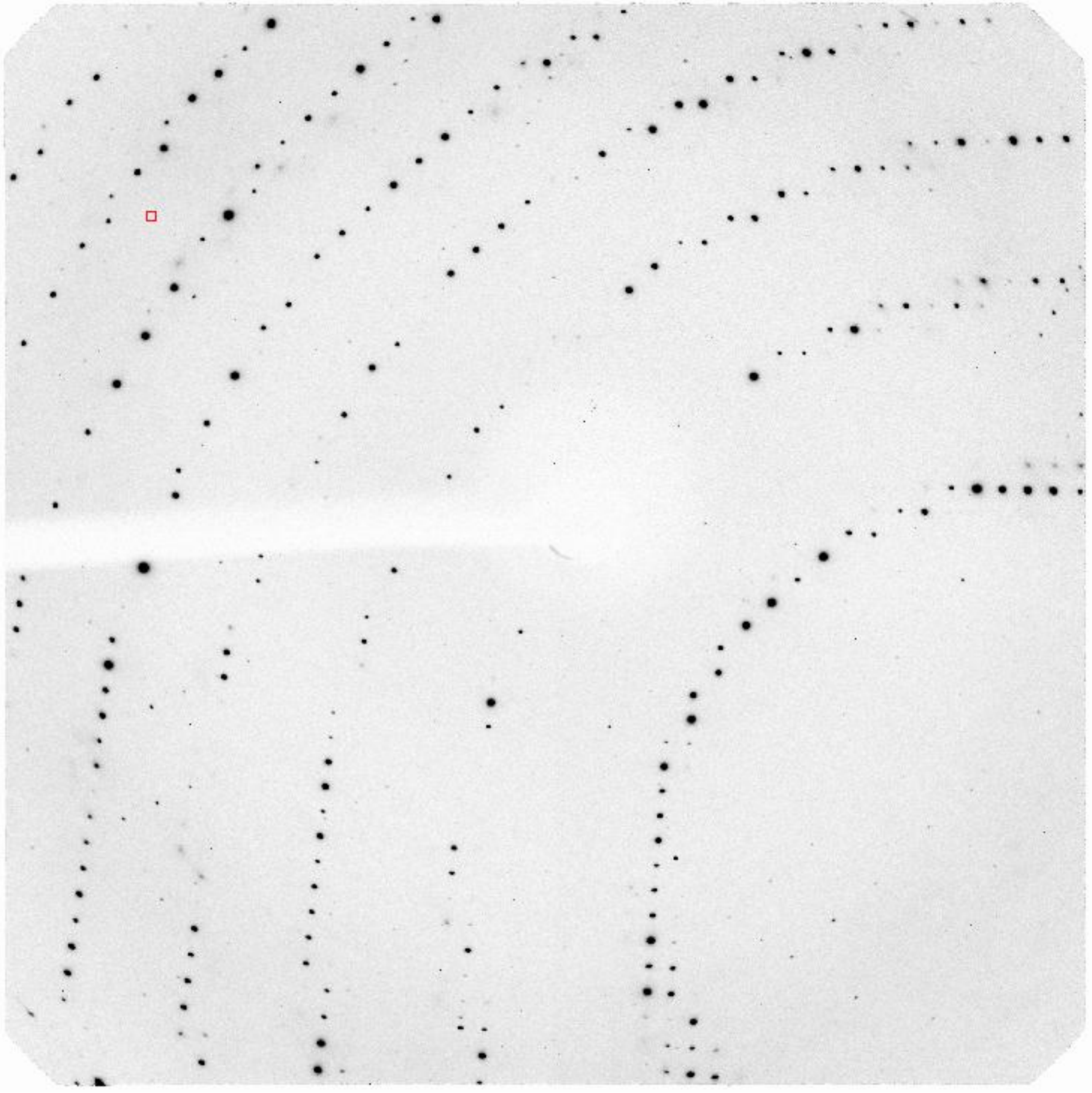

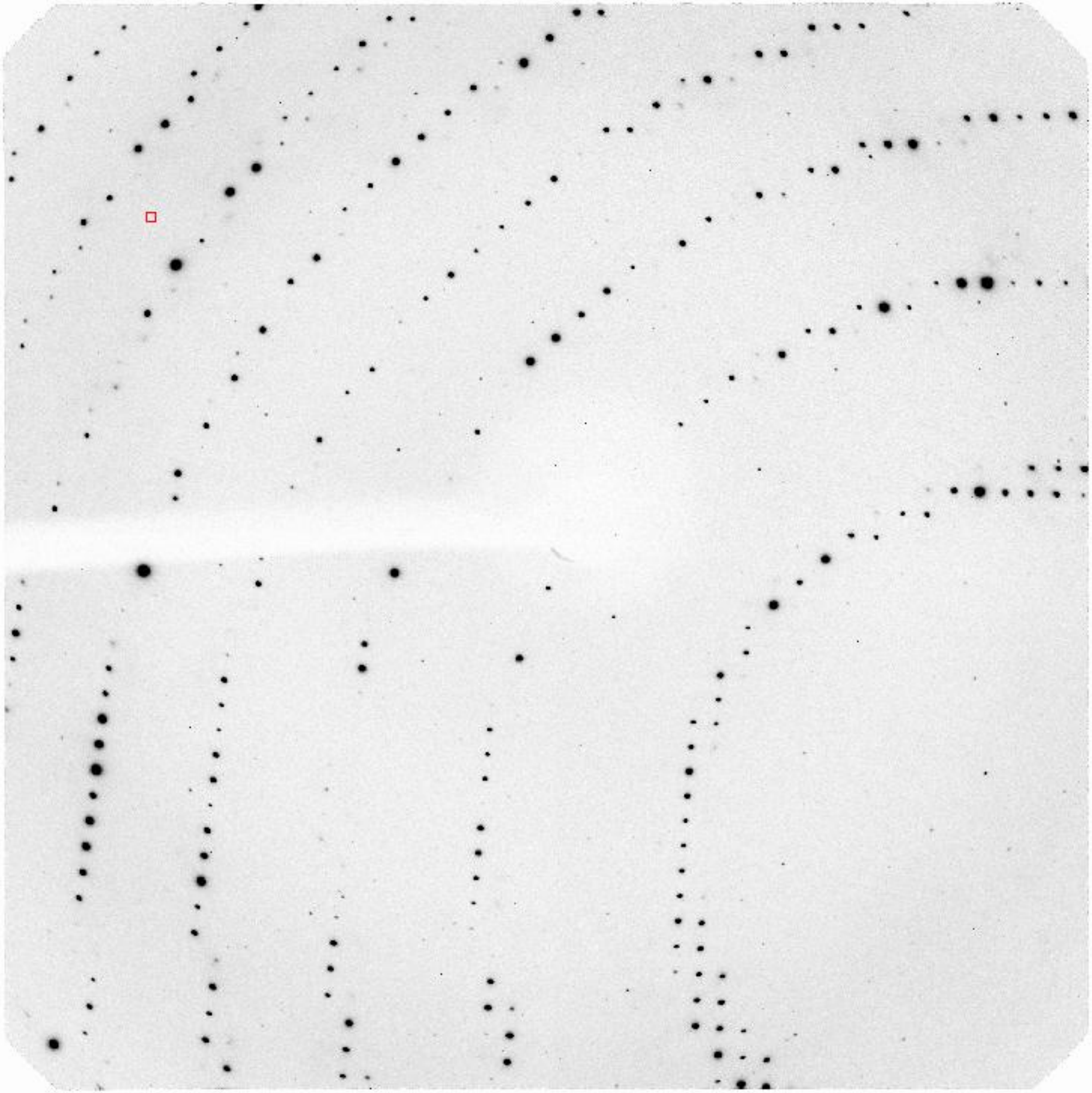

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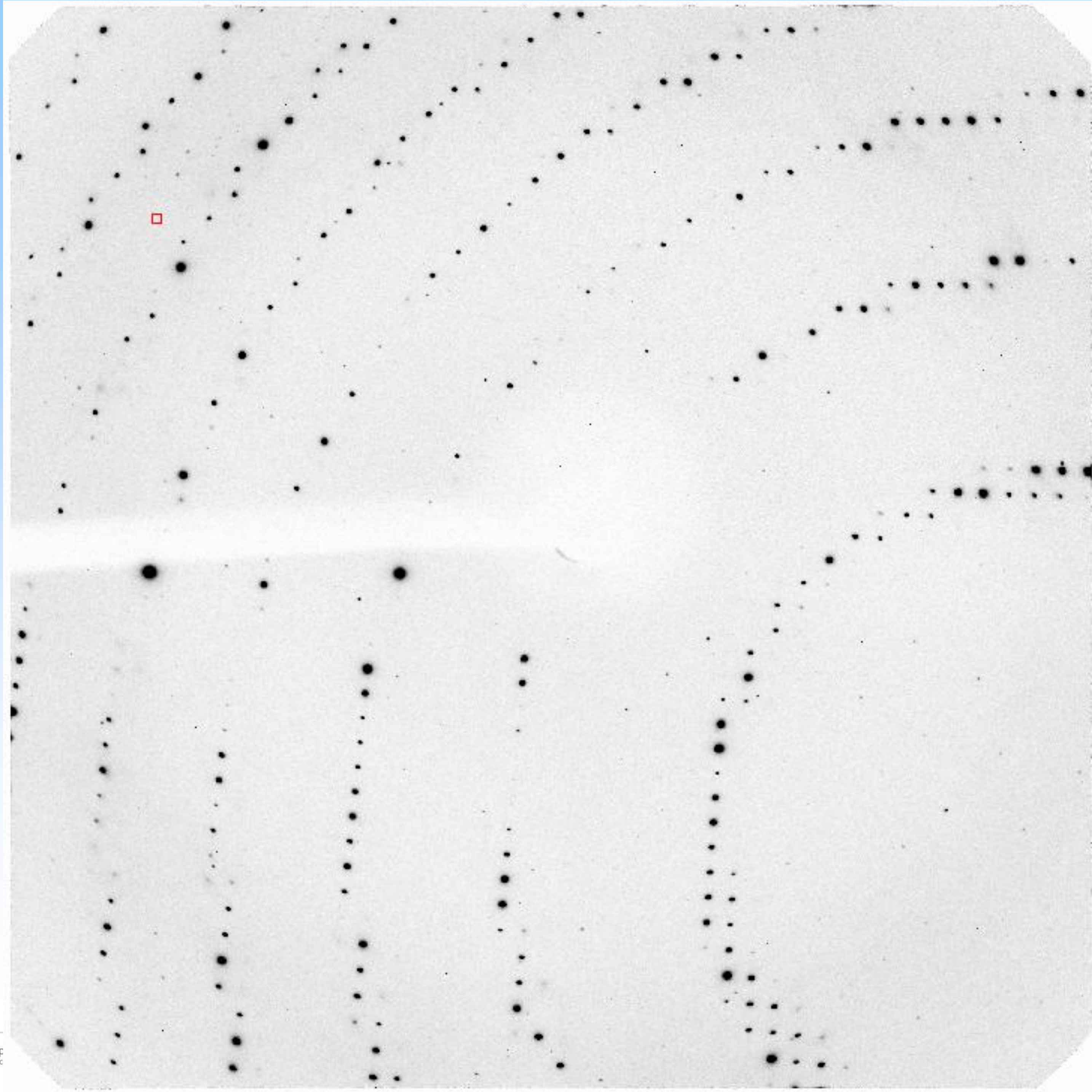

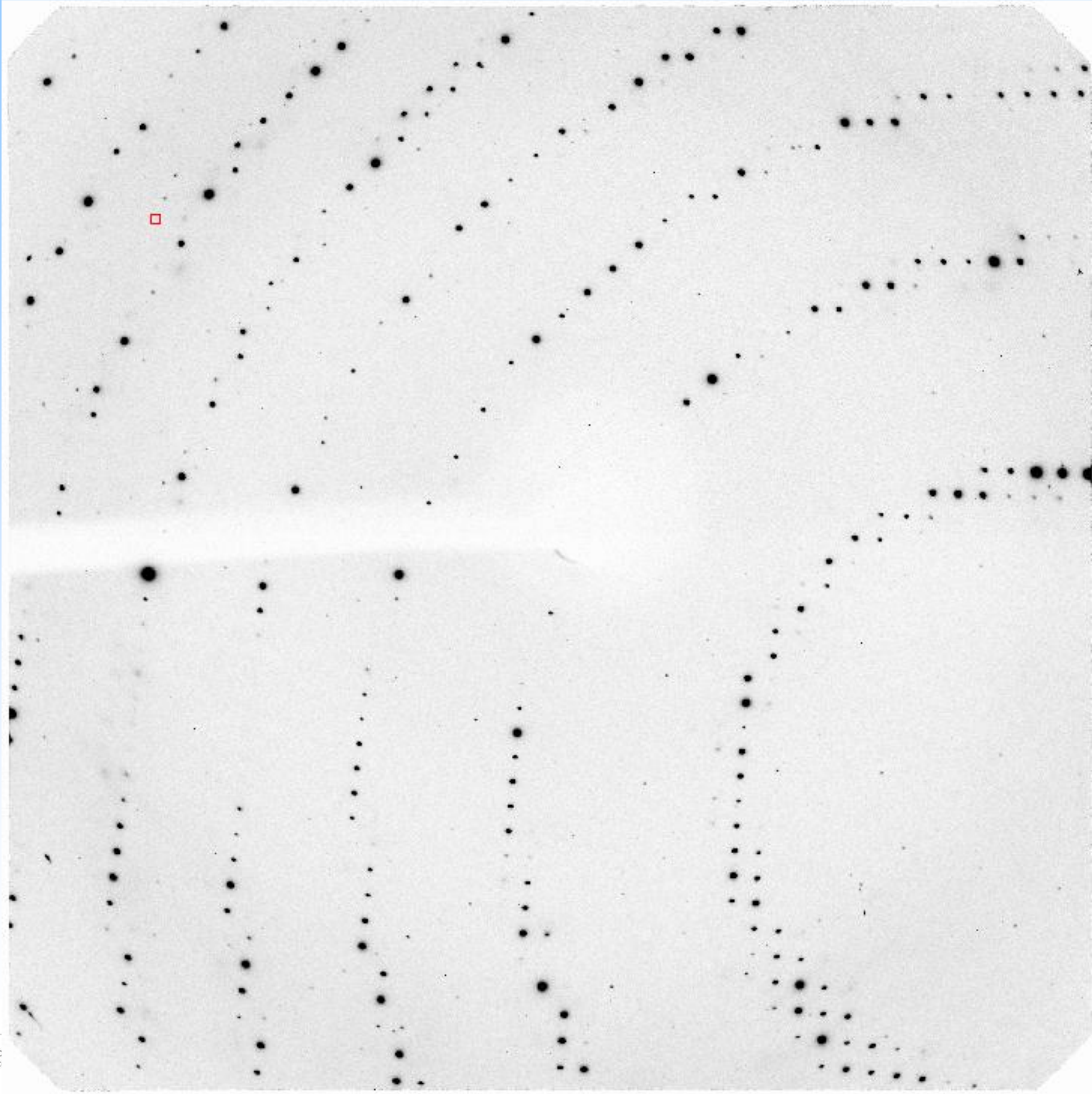

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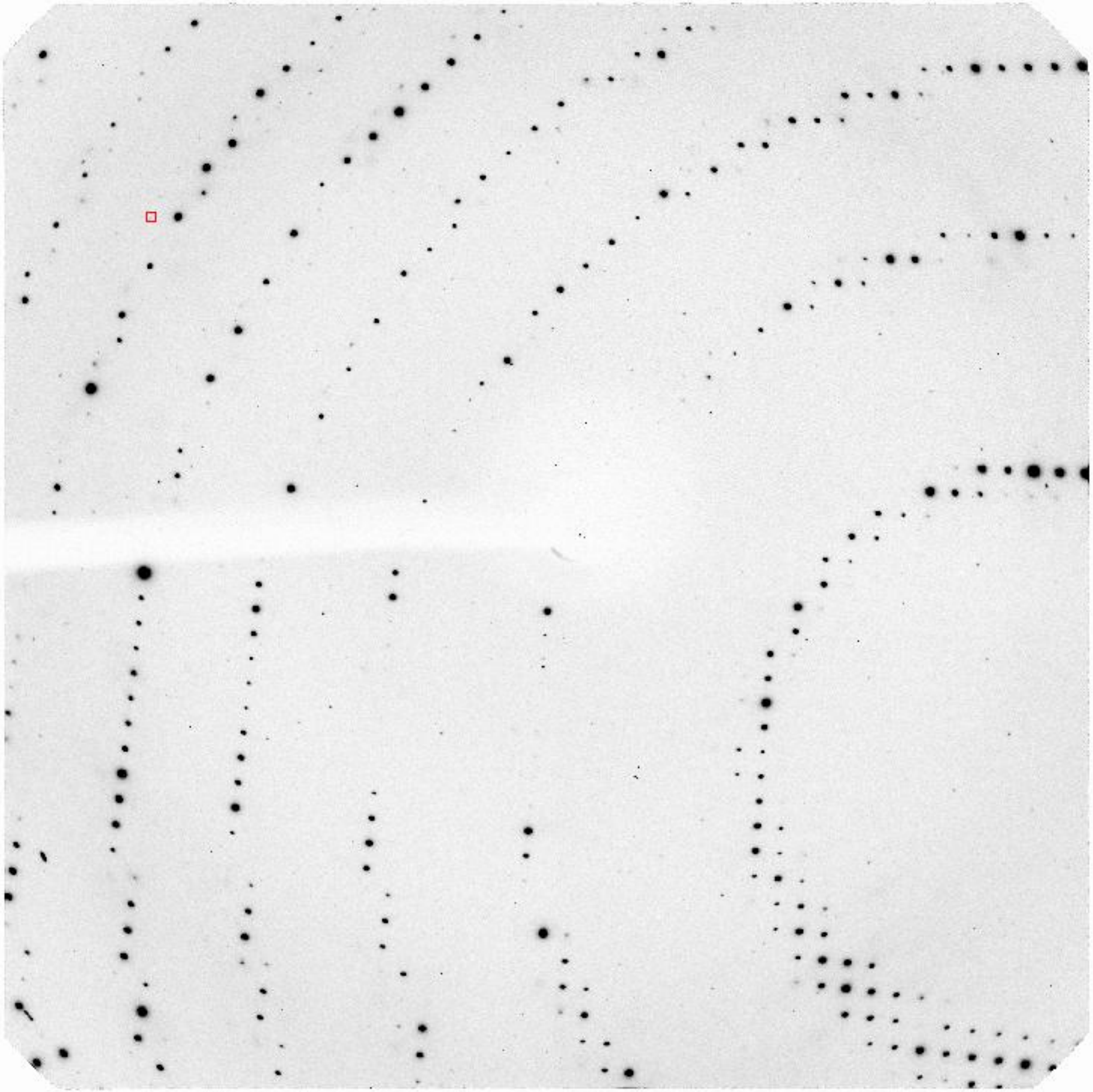

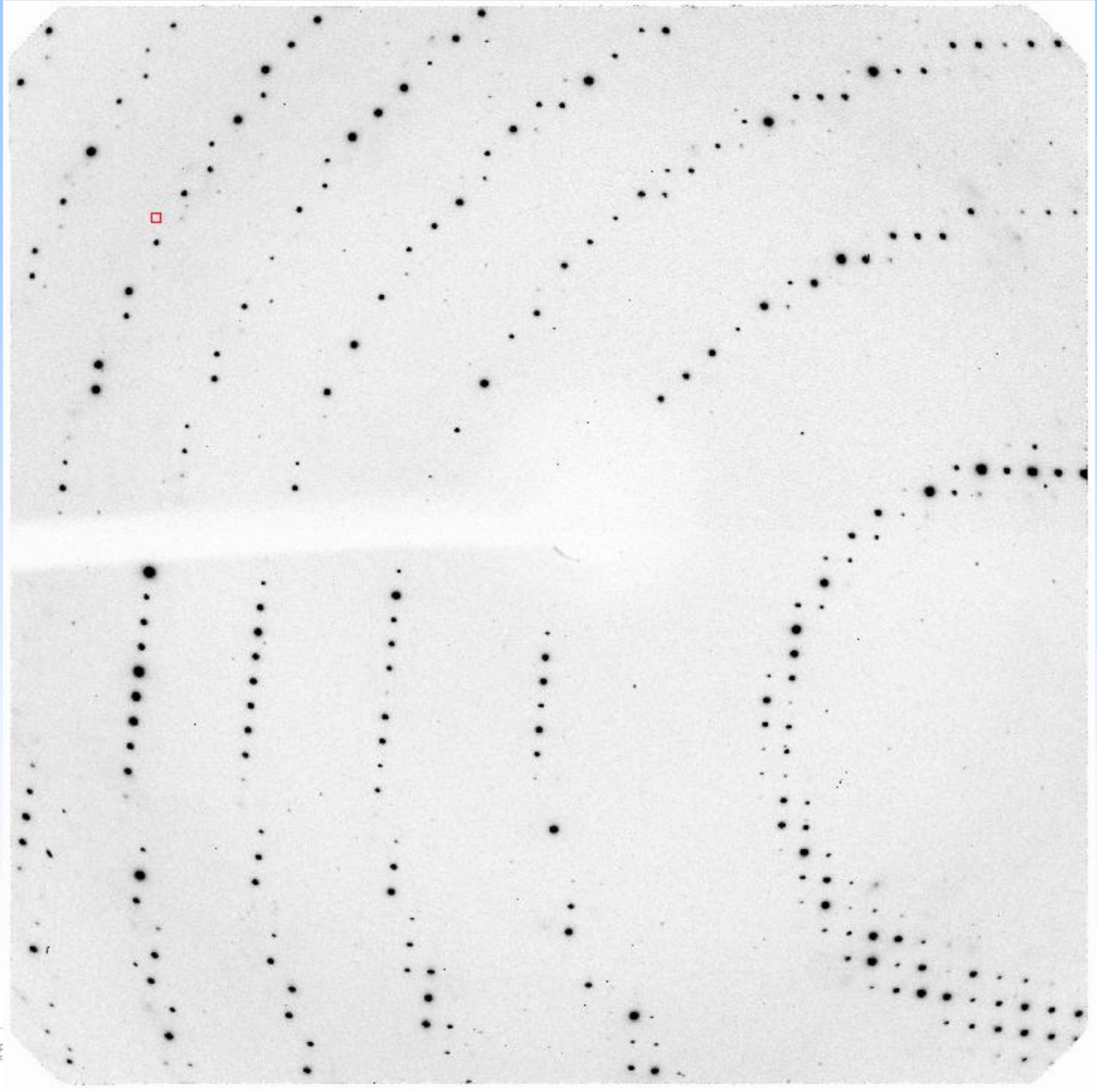

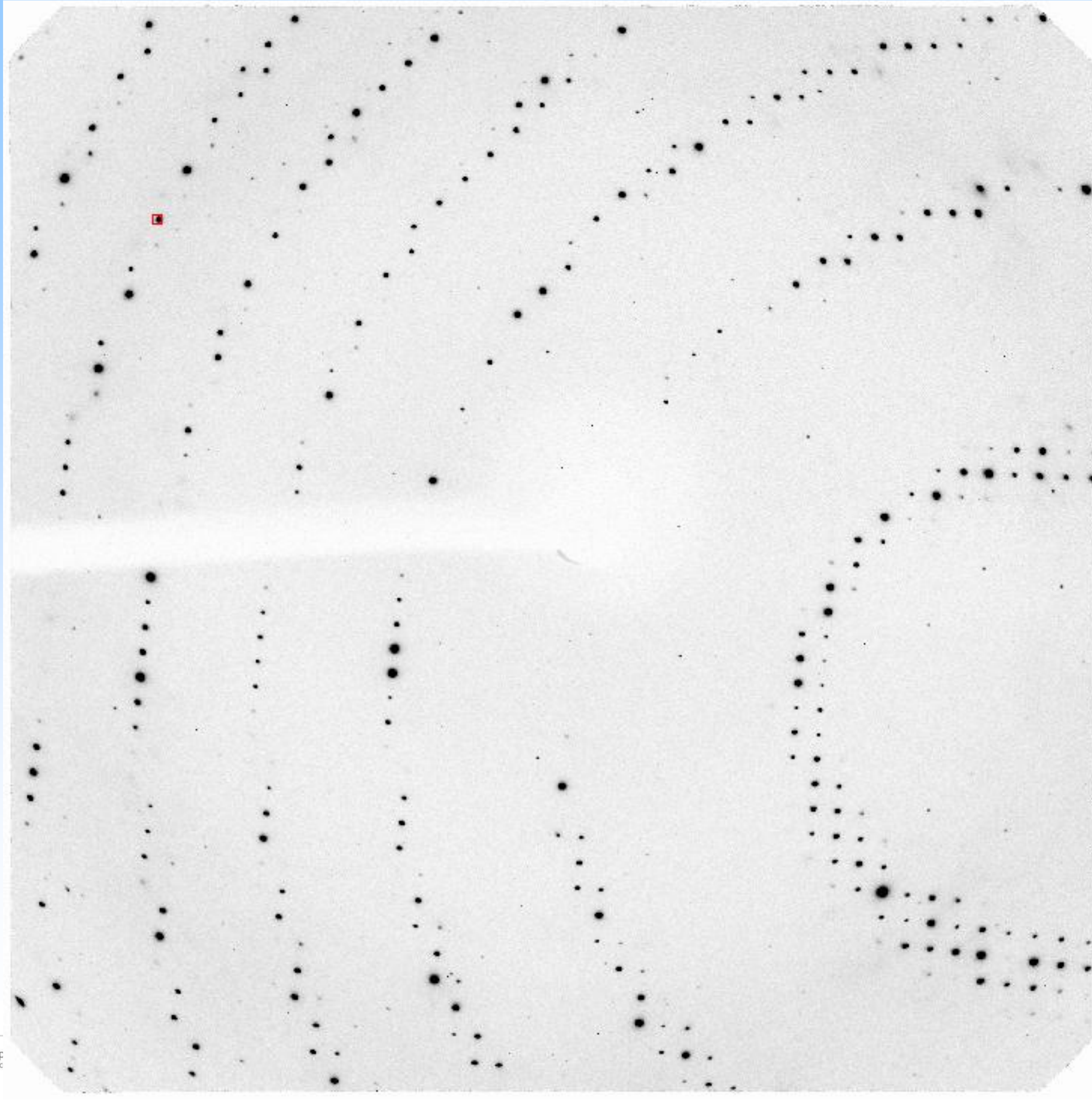

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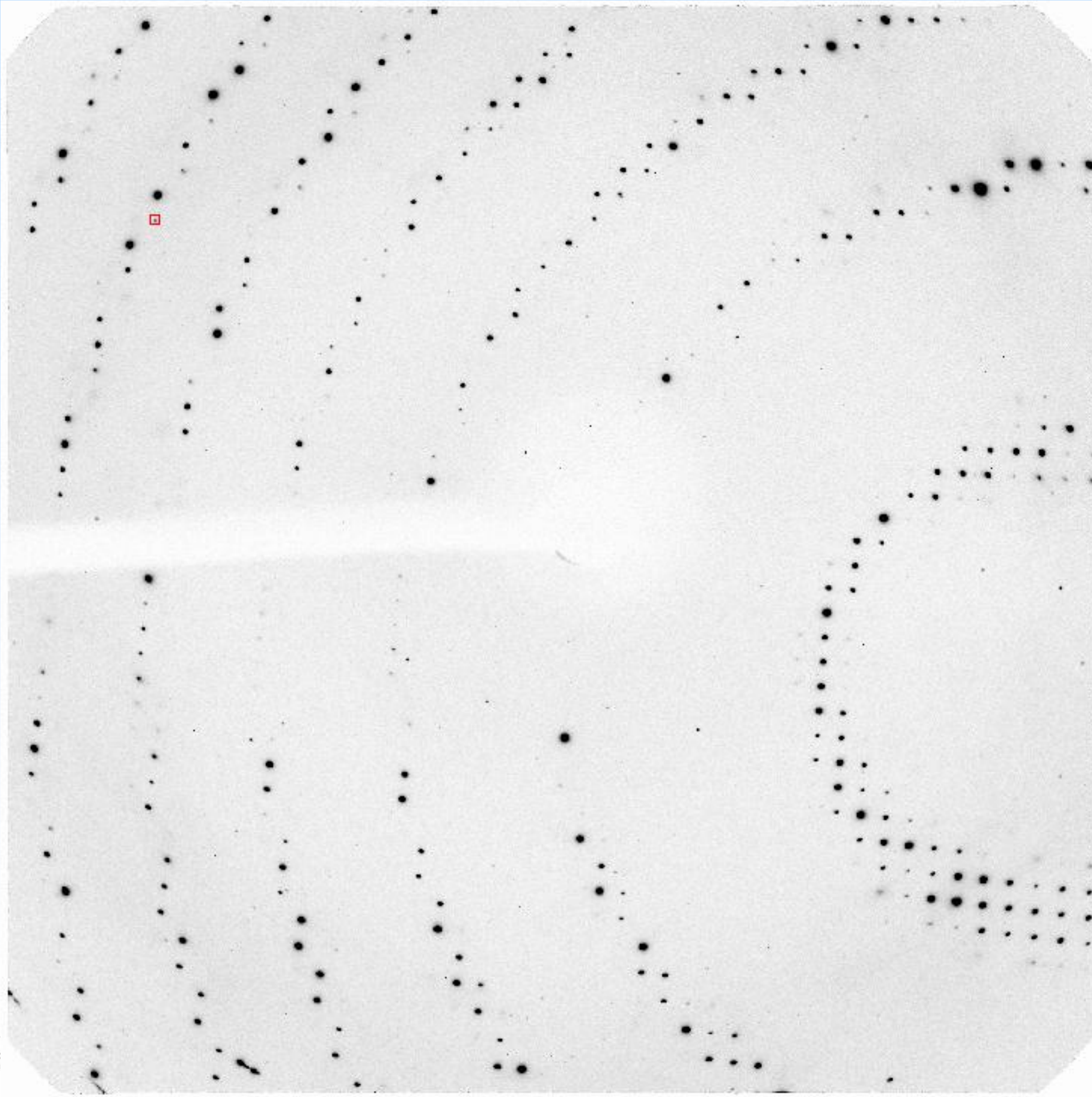

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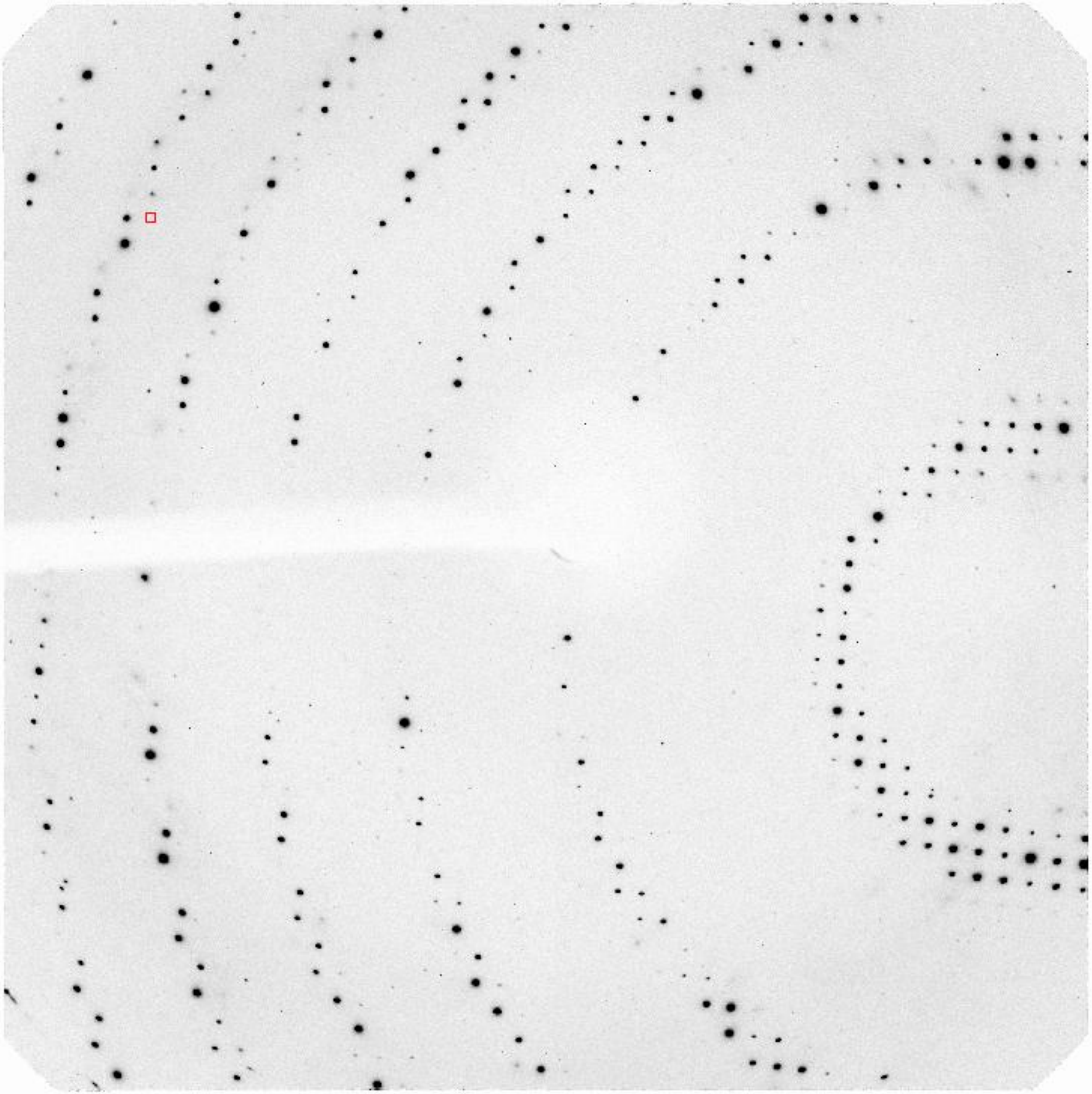

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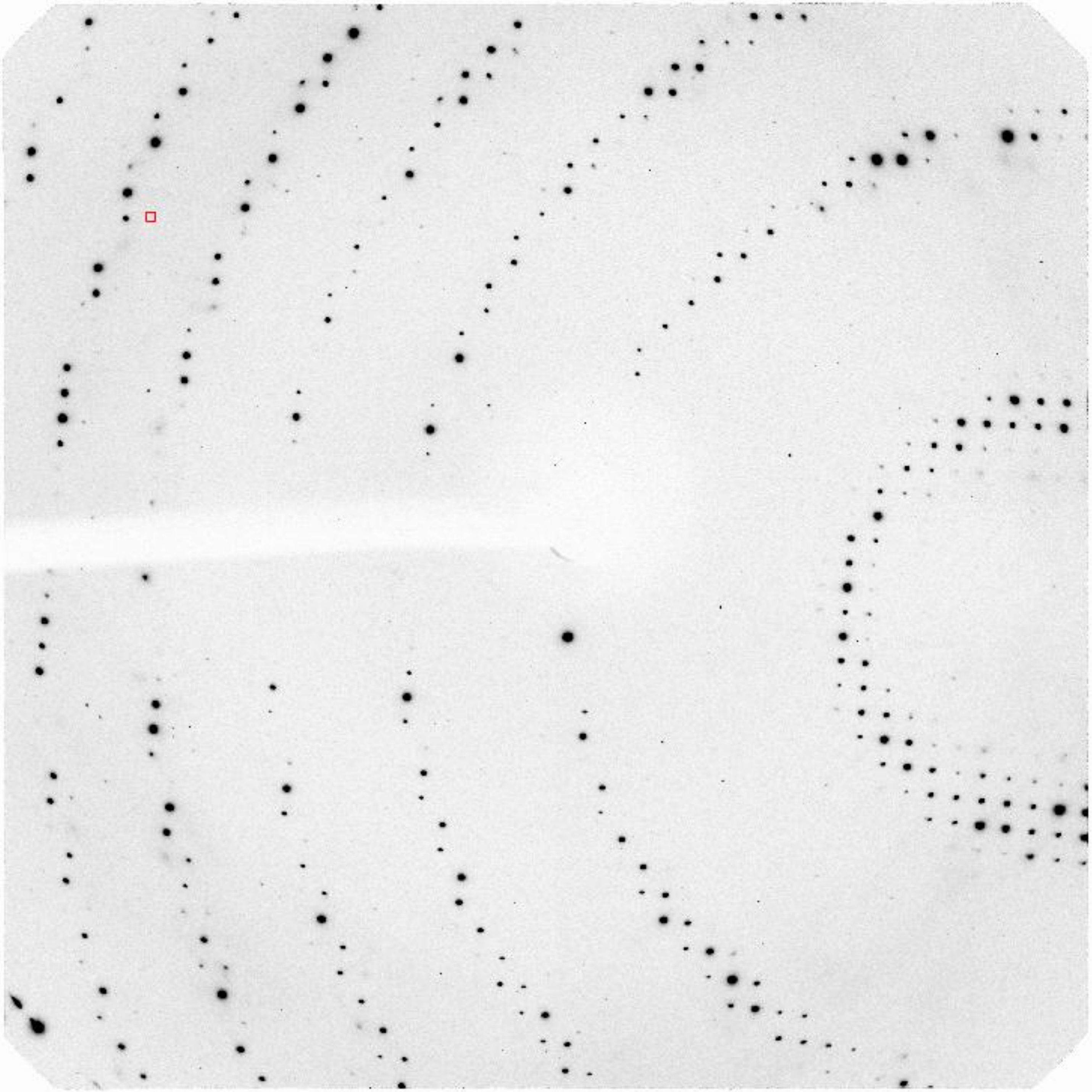

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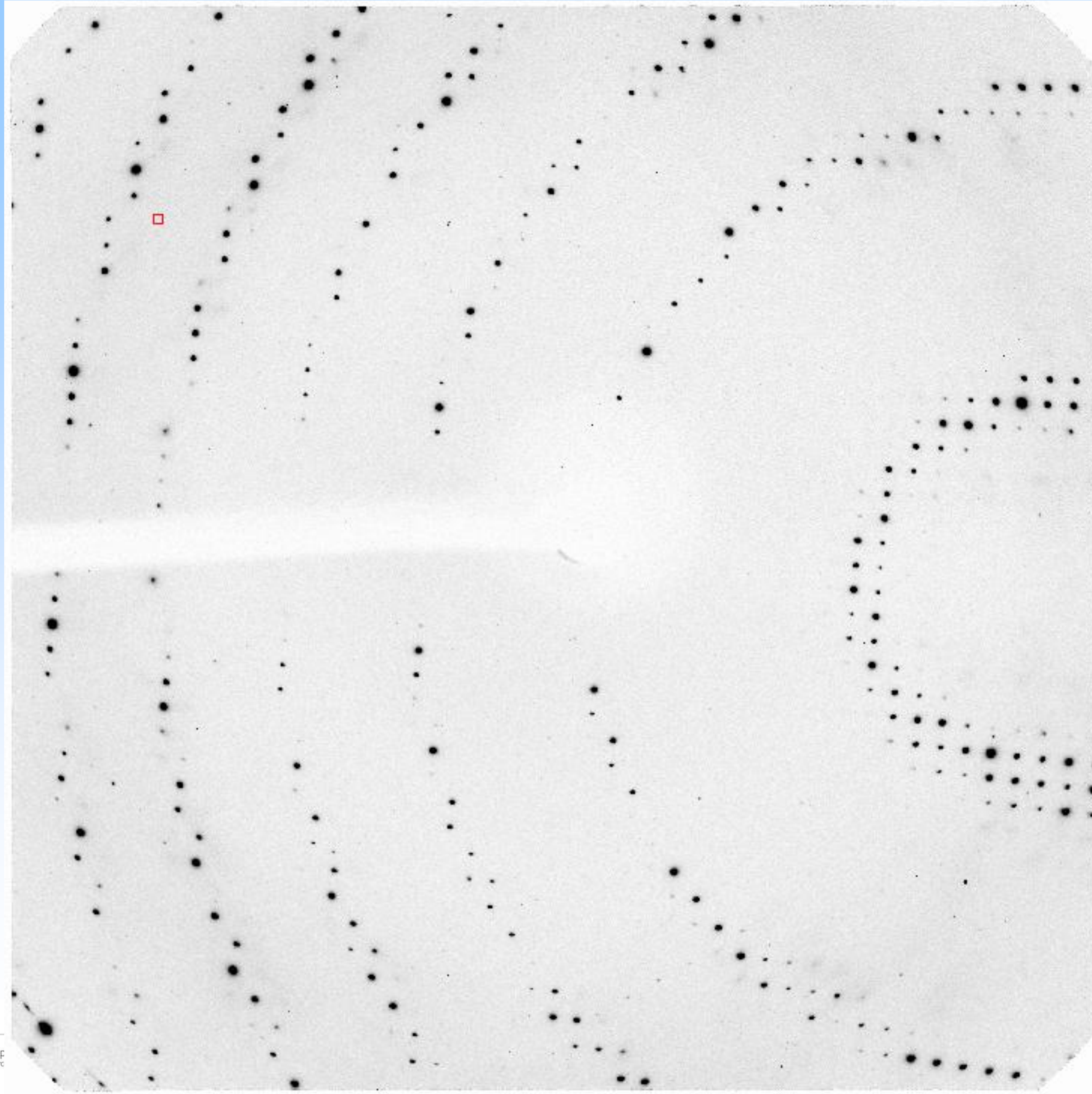

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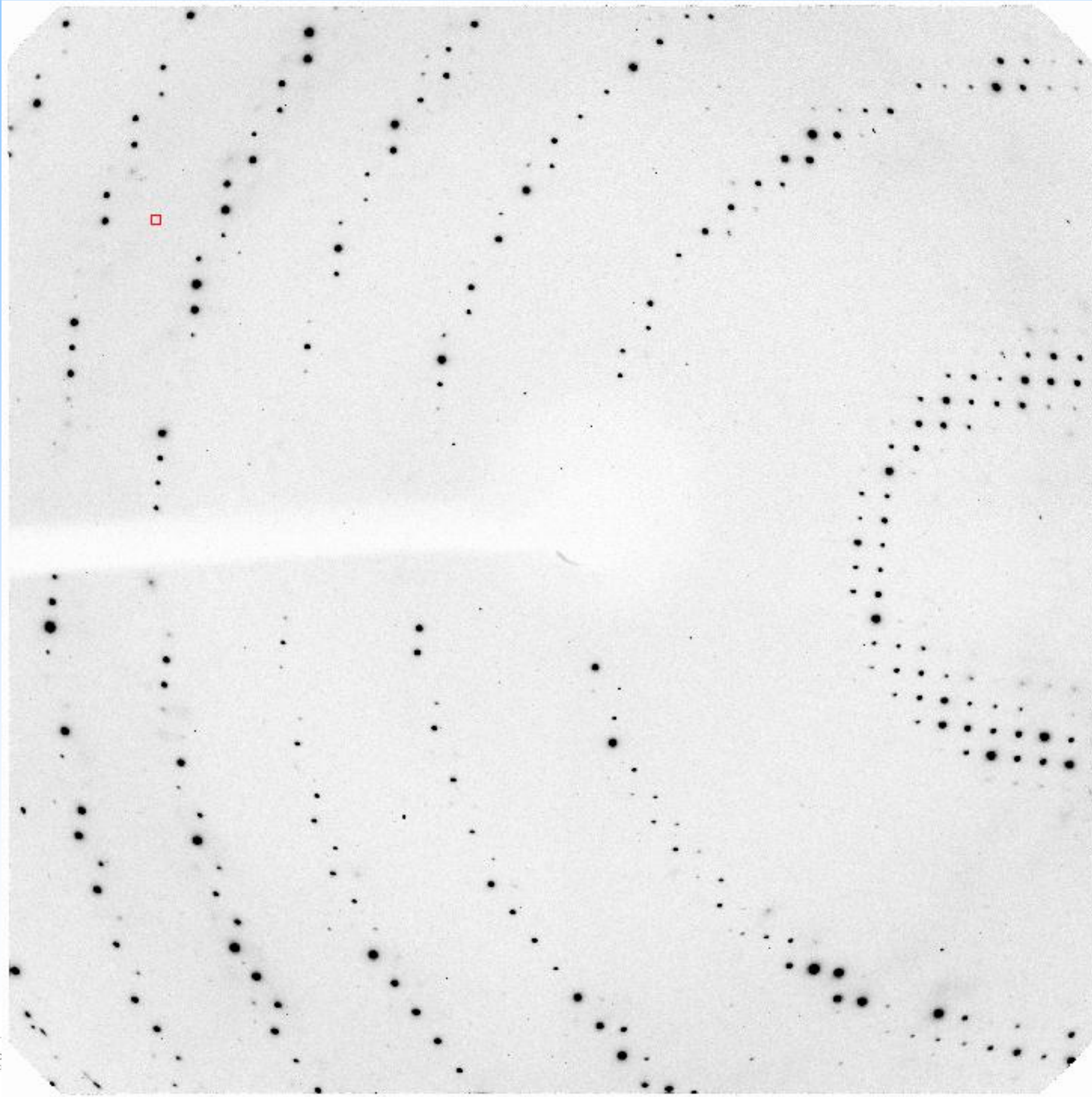

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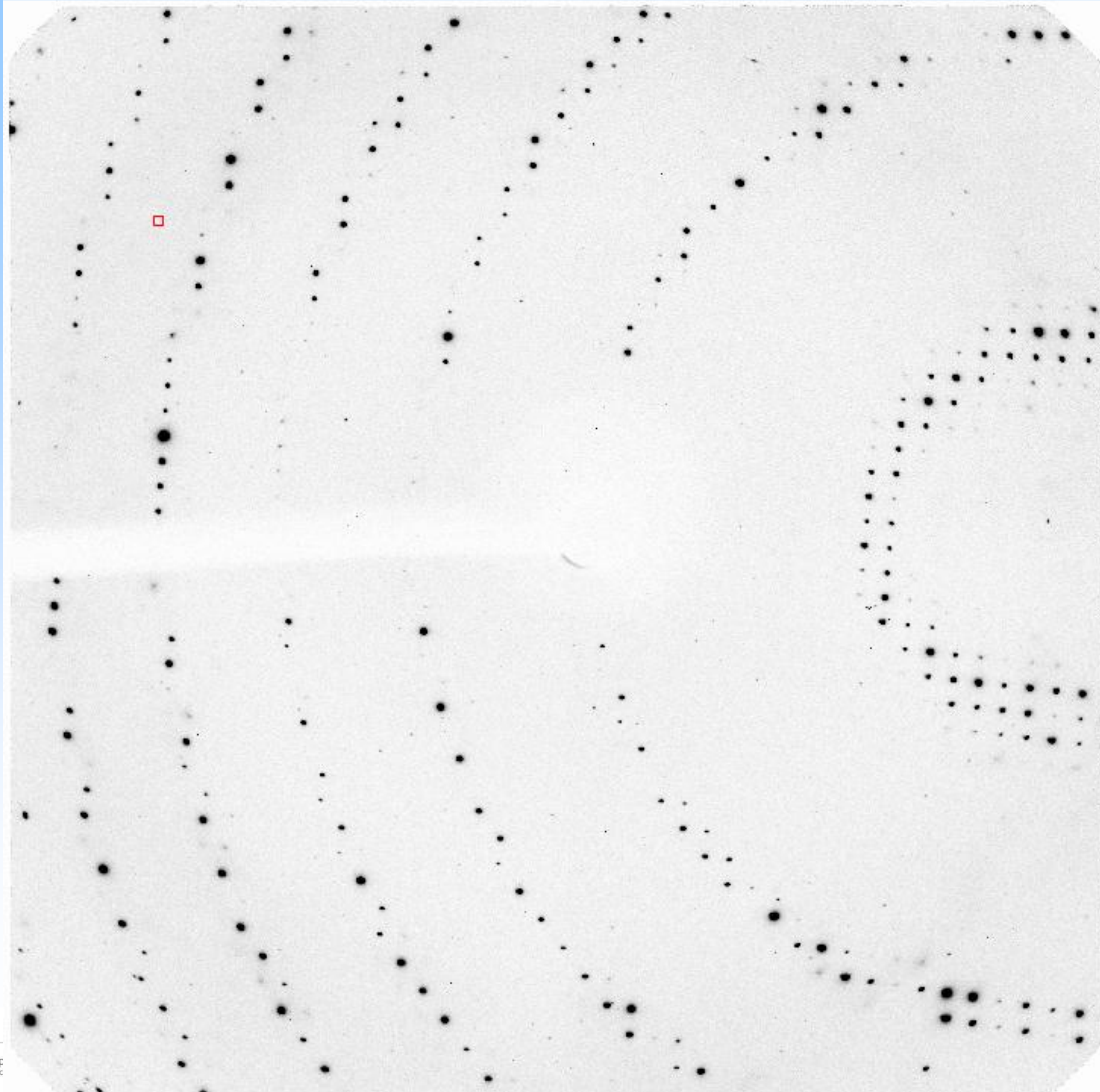

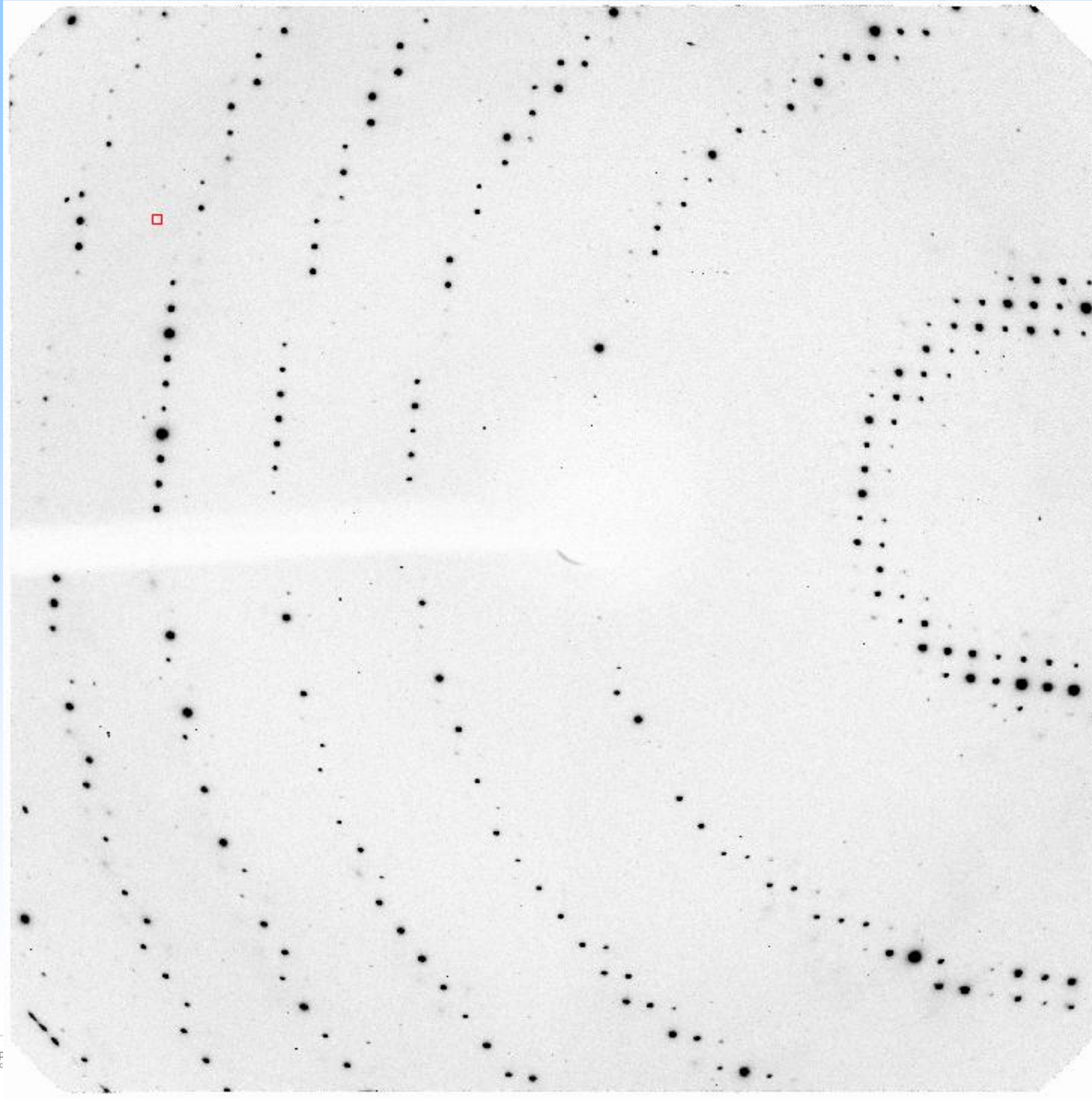

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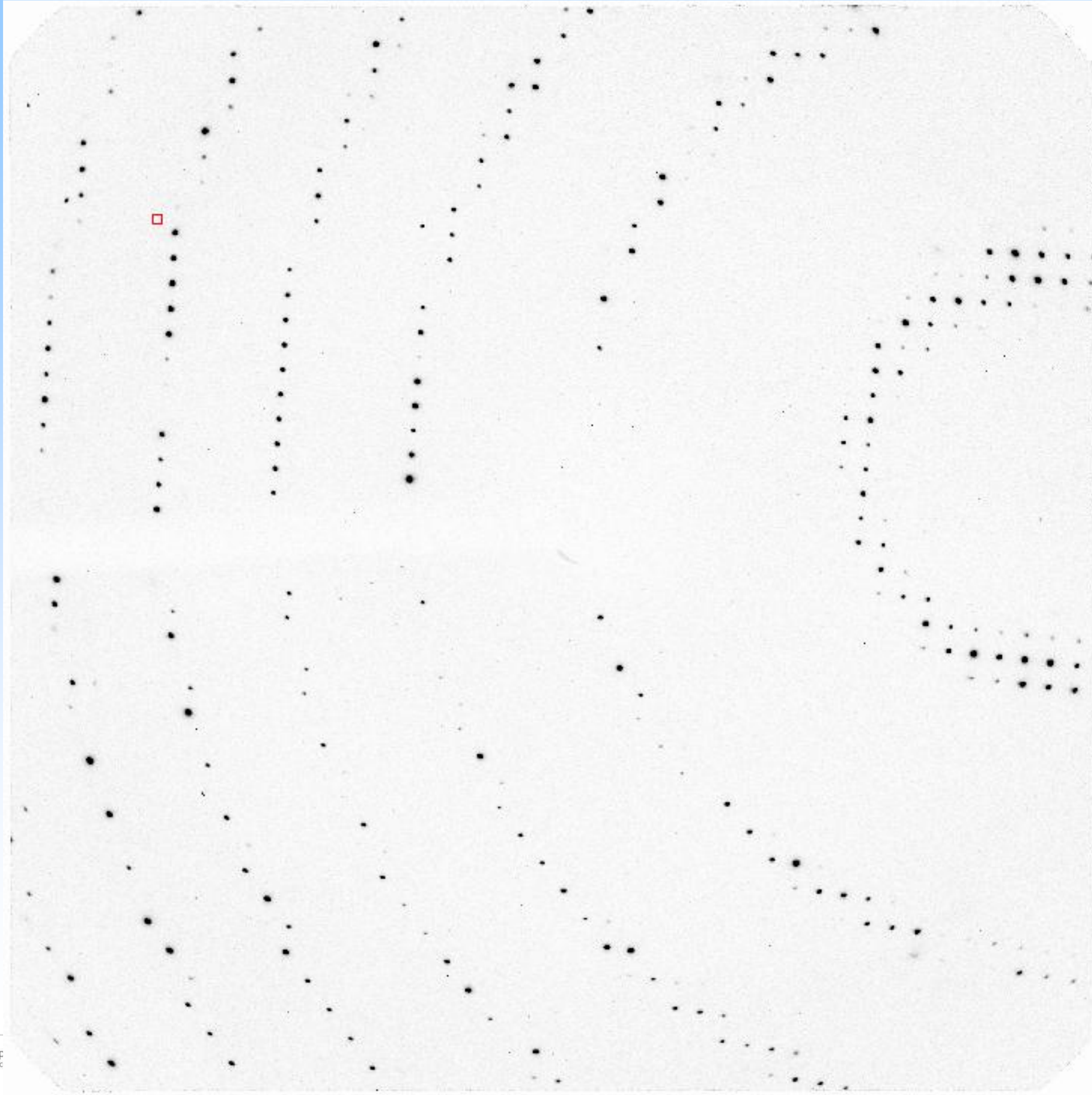
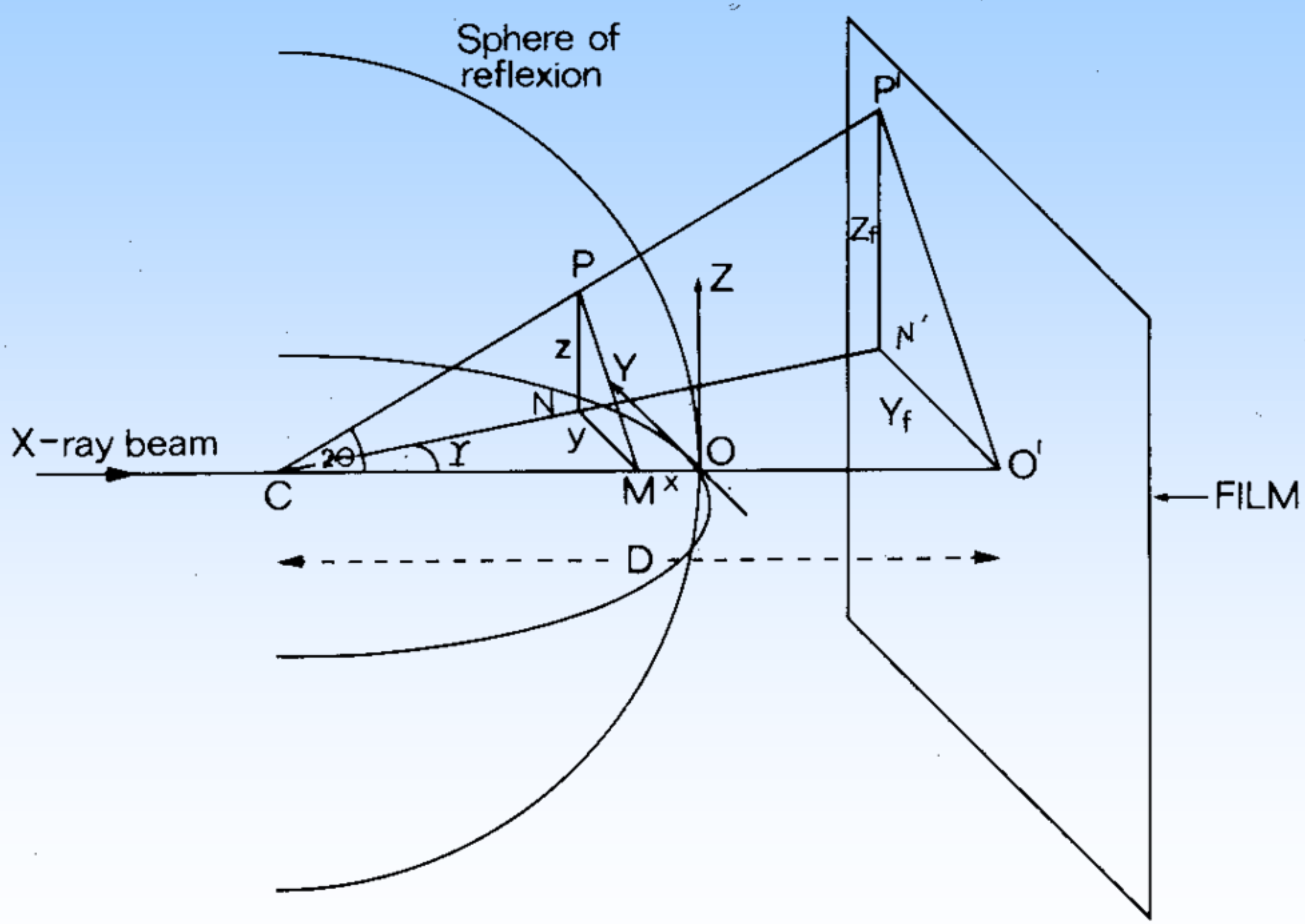


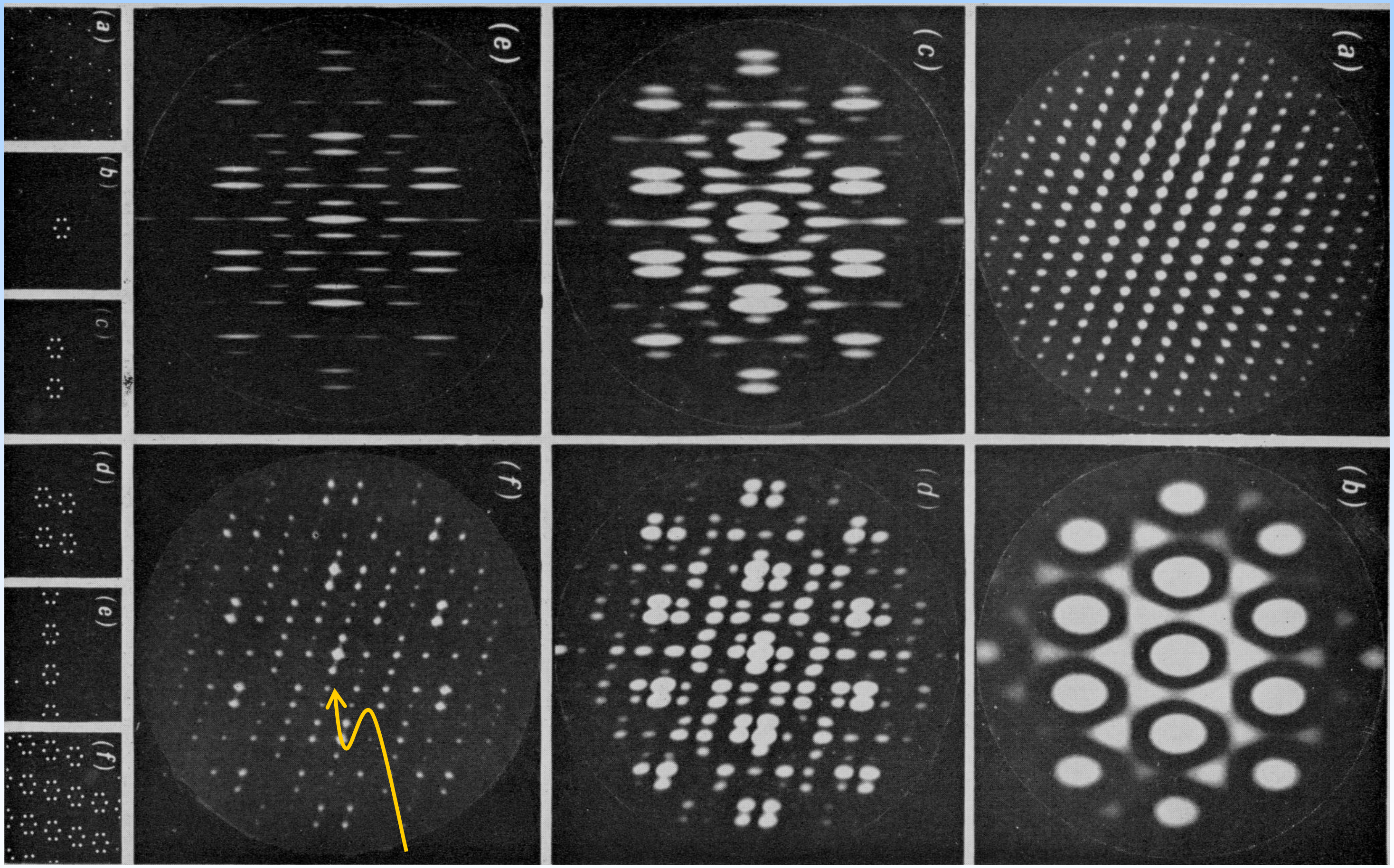
The position of the reciprocal-lattice points can be nicely related to the coordinates of reflections on the film/detector



Outline for the Lecture

- Remind you how much you already know -- lenses, crystals
- Show why crystals give diffraction spots.
- Develop the idea of "The Reciprocal Lattice"
- Give some idea how we might actually measure diffraction data
- **Show how, given a crystal, we can calculate the diffraction pattern**
- Conversely, show how to calculate the structure from the diffraction
- Describe the importance of symmetry to diffraction
- Outline the structure-solving methods -- heavy atoms and MADness

Now we use the Taylor and Lipson figures to see how the **contents** of the crystal relate to the diffraction pattern.



crystal

Coordinates of points are “indices.”

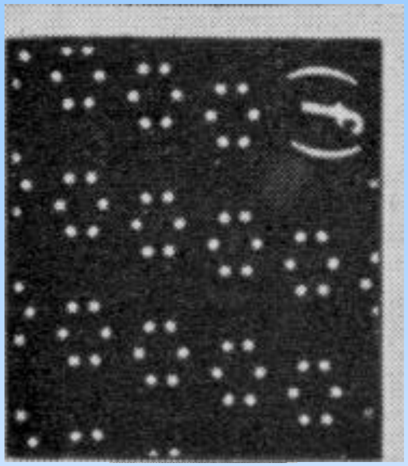
diffraction

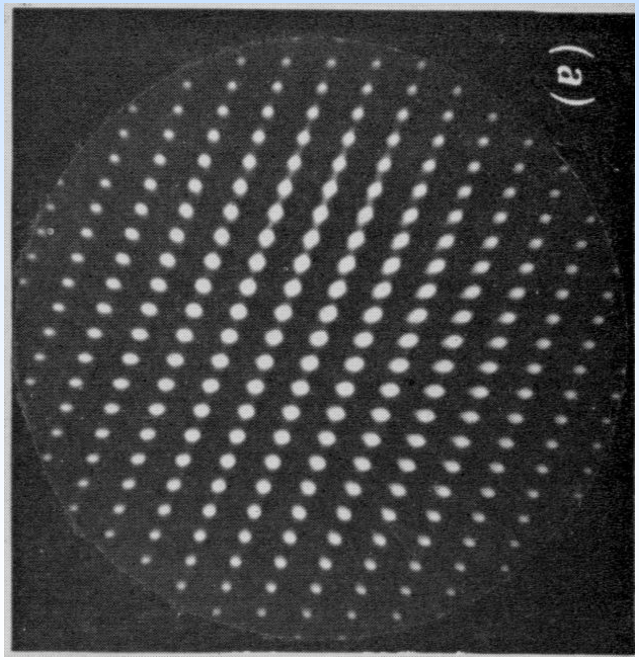
Plate 2 from Taylor and Lipson -- Optical Transforms

Notice (1) The symmetry, and (2) how the continuous diffraction pattern of one molecule (b) is “sampled” by the lattice of diffraction points.

Review: Do we understand the real/reciprocal lattice idea?

Crystal –
Real Lattice





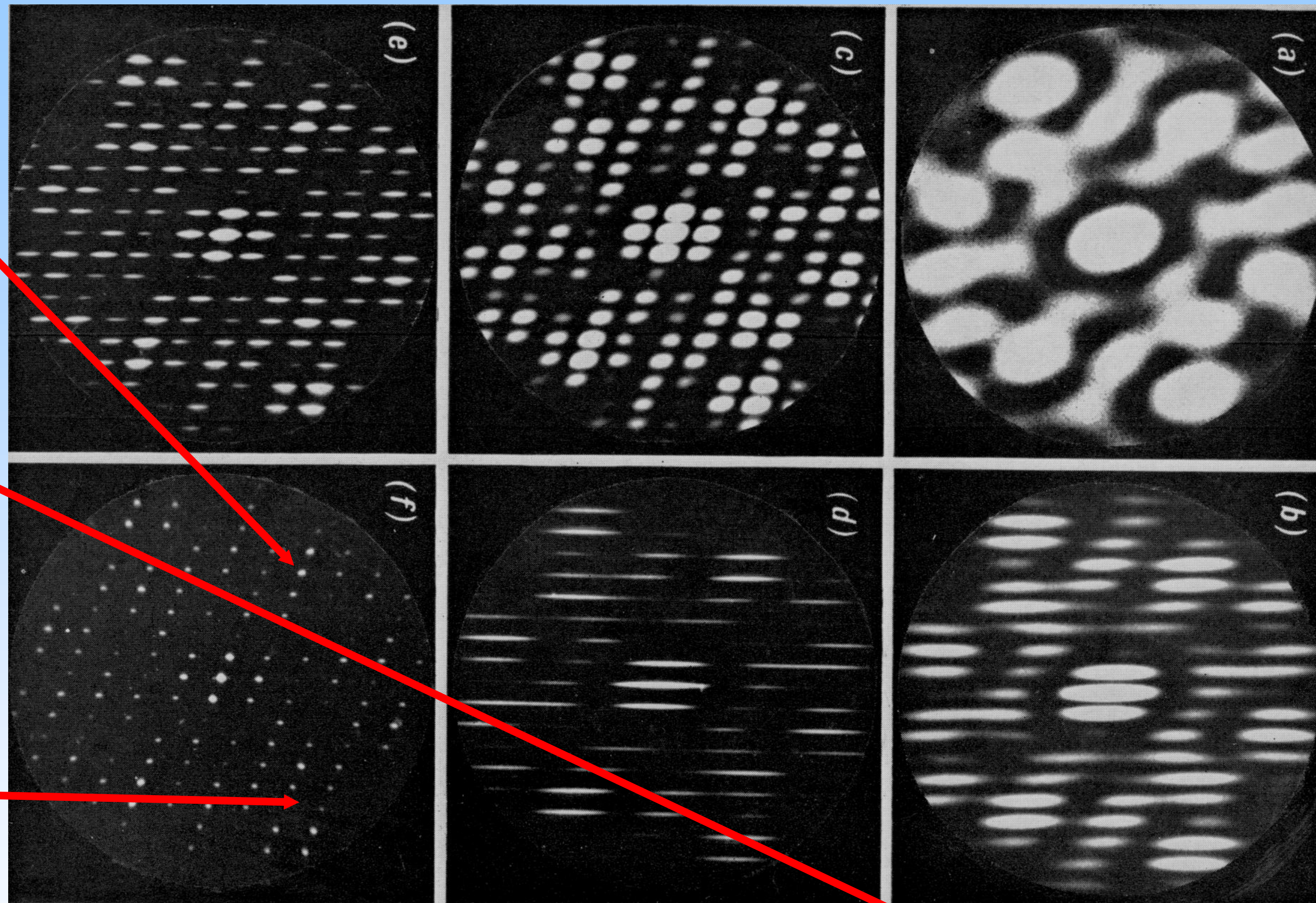
Diffraction –
Reciprocal Lattice

Confirm that the vectors perpendicular to the **Crystal-Lattice planes** are parallel to the **Reciprocal Lattice vectors**, and that the reciprocal distances make sense.

Here's another (*2D)* example with an asymmetric motif

Each spot represents the intensity of reflection from one set of planes cutting through the crystal

Note the inversion symmetry



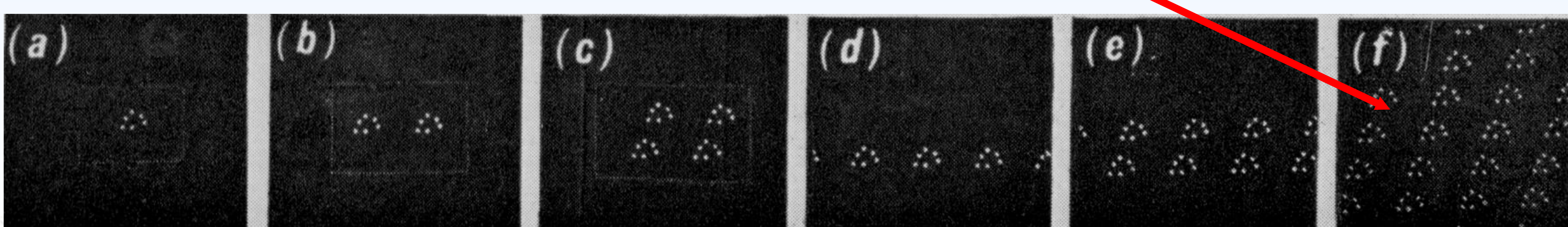
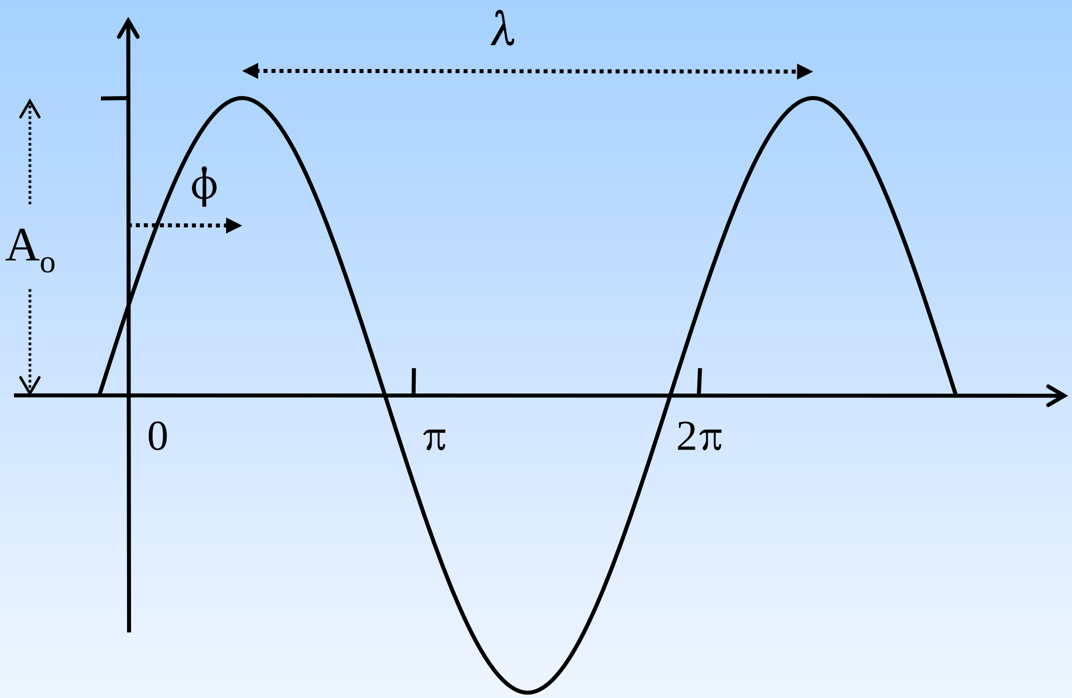
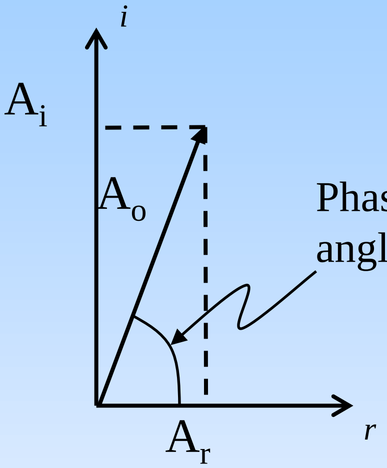


Plate 26 from Taylor and Lipson -- Optical Transforms

To calculate the structure factor we need to think of wave-like x-rays interacting with atoms. Remember that we can use an x/y graph to represent the phase and amplitude of a wave:



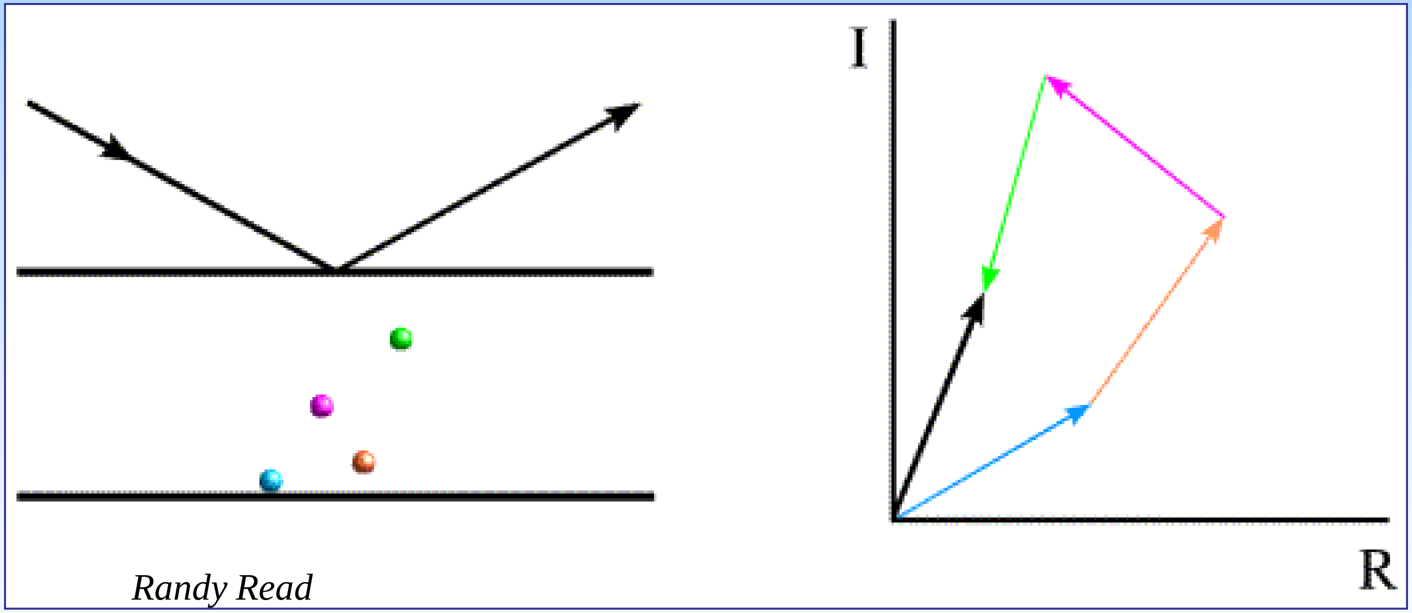


And then we describe the "wave" as a complex number:

$$\mathbf{f} = A_o\{\cos\phi + i\sin\phi\} \text{ and}$$

$$\mathbf{f} = A_o\, e^{i\phi}$$

• The amplitude of scattering depends on the number of electrons on each atom.
• The phase depends on the fractional distance it lies from the lattice plane.



Scattering from lattice planes

Atomic structure factors add as **complex numbers**, or **vectors**.

We can write an expression to describe this diffraction from atoms in a crystal

Notice that λ and the unit cell parameters are ***NOT*** *part of this.*

The scattering **amplitude** (the structure factor) for an individual atom is going to be:

The hkl describe the Bragg Planes

$$f_{hkl} = f_j \exp[2\pi i(hx_j + ky_j + lz_j)]$$

The scattering power of the atom, ~ the number of electrons

The 2π and the fractional coordinates x_i take care of the phase angle

And the **structure factor** for a crystal of atoms will be:

$$F_{hkl} = \sum_{\text{atoms}} f_j \exp[2\pi i(hx_j + ky_j + lz_j)]$$

The strength of scattering from each atom

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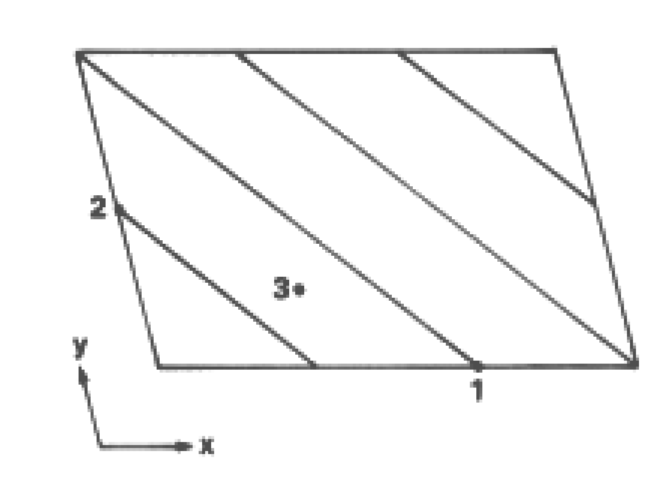
Does this expression for the Structure Factor make sense?

Try it with an example: a crystal with three atoms. What are the phases of scattering from each atom? Use this

$$f_{hkl} = f_j \exp[2\pi i(hx_j + ky_j + lz_j)]$$



For these planes, (h, k) = (3, 2)

For atom 1. x, y = 2/3, 0: So $2\pi(hx + ky) = 2\pi(3 \times 2/3 + 2 \times 0) = 4\pi = 0$
The atom is on the plane, so this makes sense.

For atom 2. x, y = 0, 1/2: So $2\pi(hx + ky) = 2\pi(3 \times 0 + 2 \times 1/2) = 2\pi = 0$
Again, the atom is on the plane, so this makes sense.

For atom 3. x, y = 1/3, 1/4: So $2\pi(hx + ky) = 2\pi(3 \times 1/3 + 2 \times 1/4) = 3\pi = \pi$
The atom lies half-way between two planes, so this makes sense.

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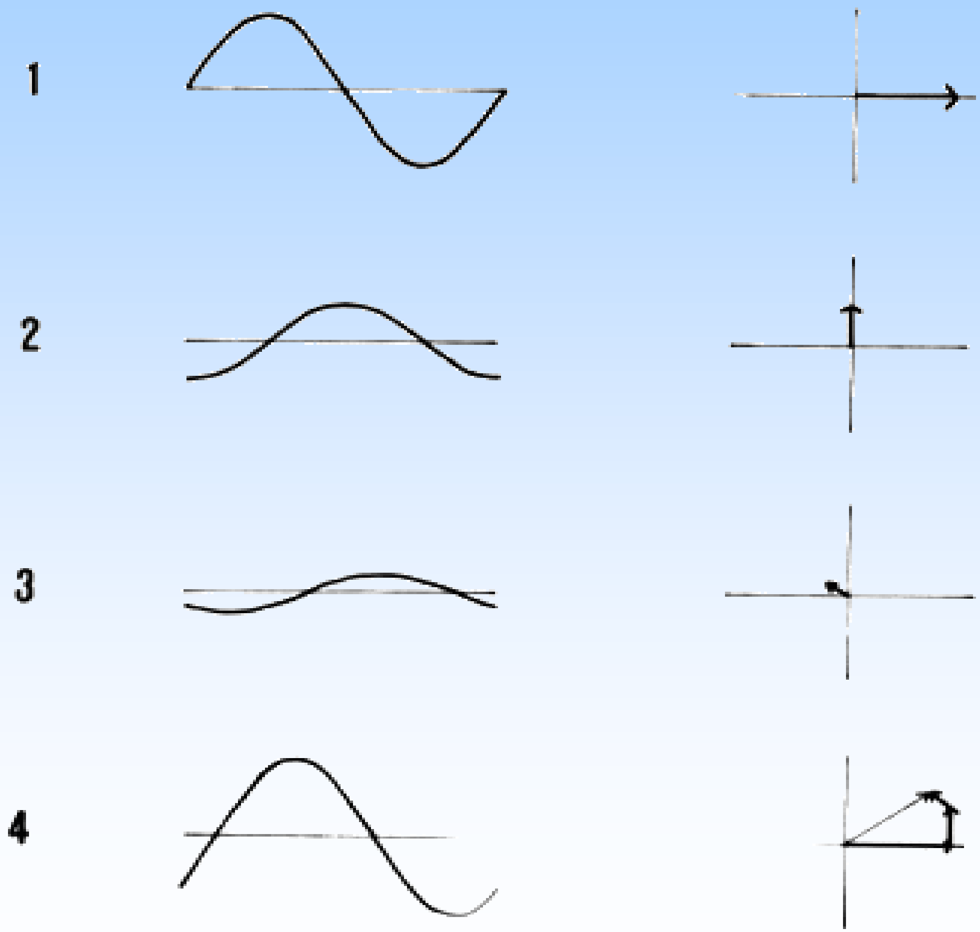
We can see how the structure factors from individual atoms add up.

Wave	Complex Vector	Complex number



$f_1 = 1 + 0i$

$f_2 = 0 + 0.5i$

$f_3 = -0.2 + 0.2i$

$f_{sum} = 0.8 + 0.7i$

See also:

http://www.ysbl.york.ac.uk/~cowtan/

sfapplet/sfintro.html	Structure Factor Tutorial
fourier/fourier.html	Book of Fourier

Outline for the Lecture

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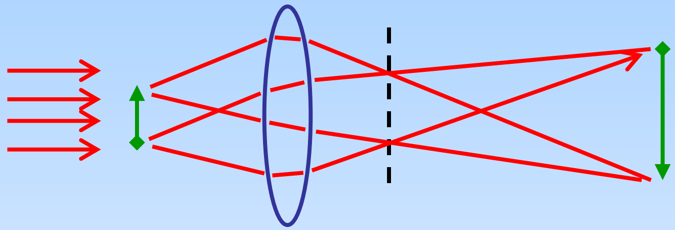
- Show why crystals give diffraction spots.
- Develop the idea of "The Reciprocal Lattice"
- Give some idea how we might actually measure diffraction data
- Show how, given a crystal, we can calculate the diffraction pattern
- **Conversely, show how to calculate the structure from the diffraction**
- Describe the importance of symmetry to diffraction
- Outline the structure-solving methods -- heavy atoms and MADness

Q: How do we perform the second interference step in the functioning of the lens -- to reconstruct the image of the original object?

A: We will have to calculate it.



Q: How will we represent that object?

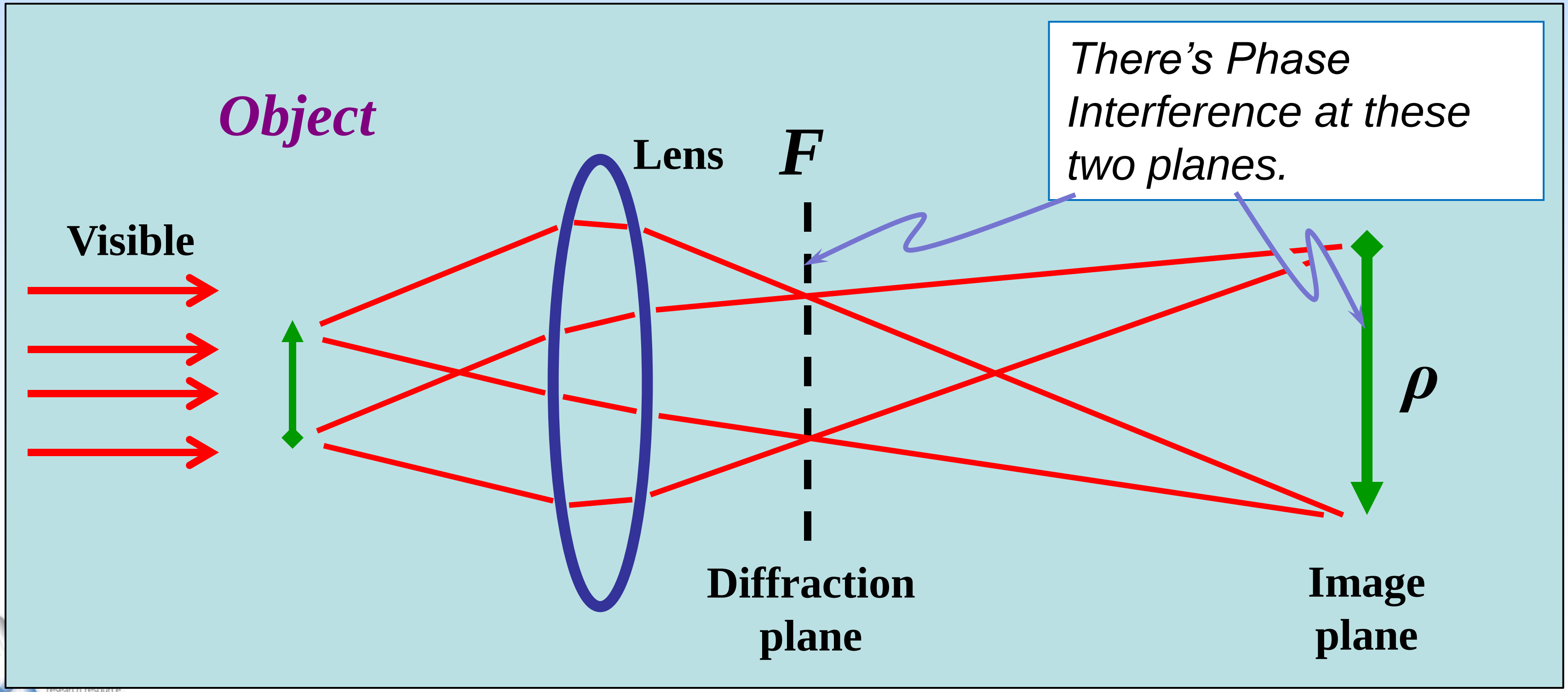
A: The x-rays are scattered from electrons in the atoms of the crystal.

Therefore: for us, the "image" is going to be a representation of the electron density.

The structure factor and the electron density function are Fourier inverses of one another

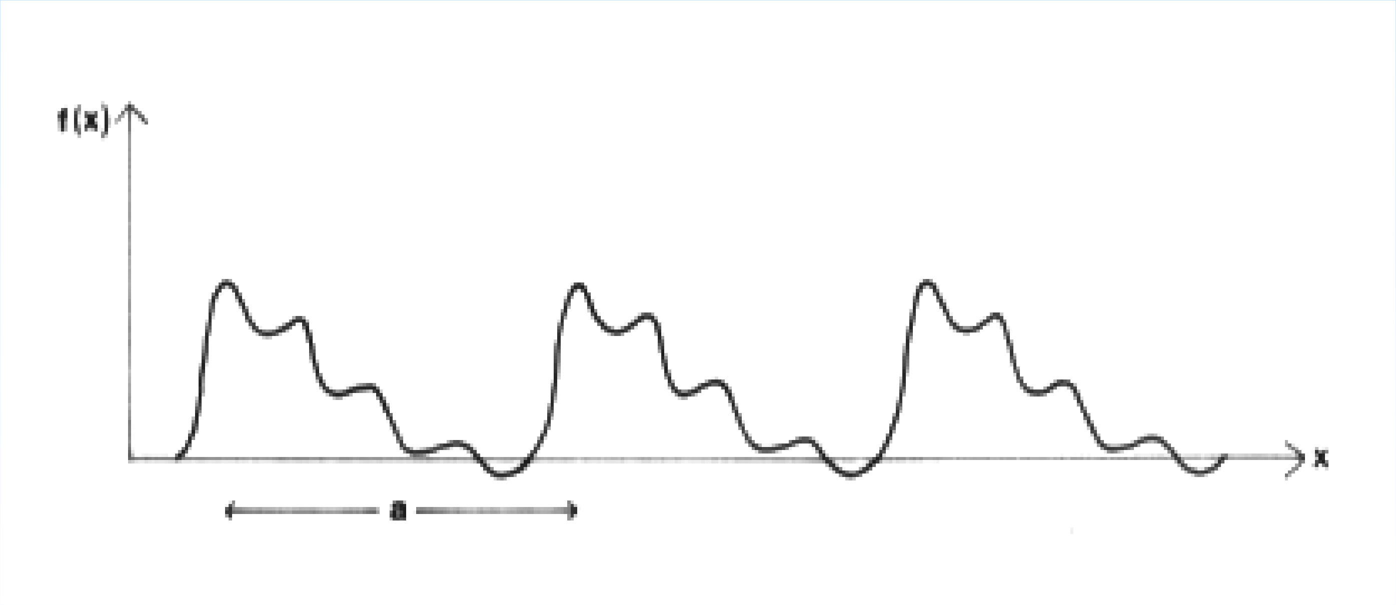
$$F_{hkl} = \int_V \rho(x,y,z) \exp[+2\pi i(hx + ky + lz)]\, dV$$

$$\rho(x,y,z) = \frac{1}{V} \sum_{h=-\infty}^{\infty} \sum_{k} \sum_{l} F_{hkl} \exp[-2\pi i(hx + ky + lz)]$$



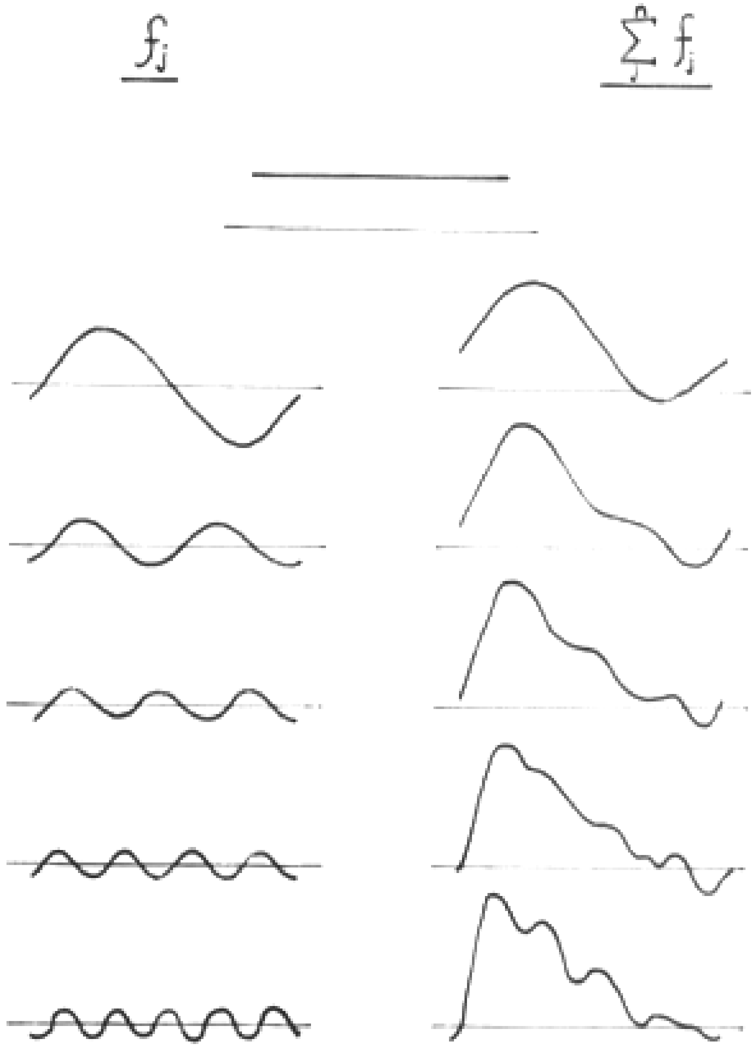
How does Fourier synthesis work?

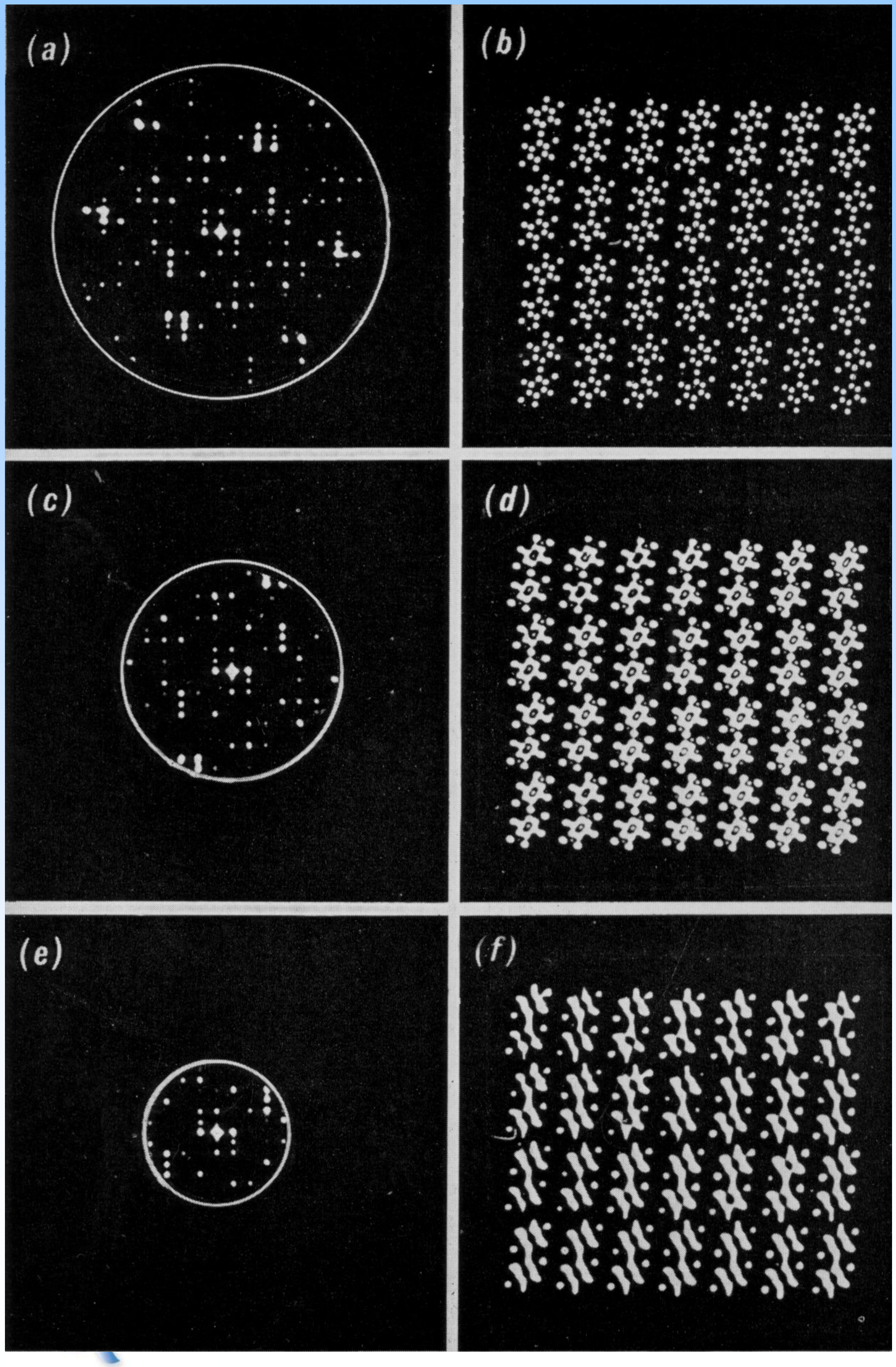
Can we produce a trial structure and see how waves can be summed to give this structure back?



In the **Fourier Synthesis**, just a few waves suffice to give a reasonable approximation to the original pattern

n	A	ϕ
0	1.00	0
1	1.21	0.6π
2	0.46	0.8π
3	0.32	0.9π
4	0.26	0.8π
5	0.29	1.3π





What is the concept of "resolution?"

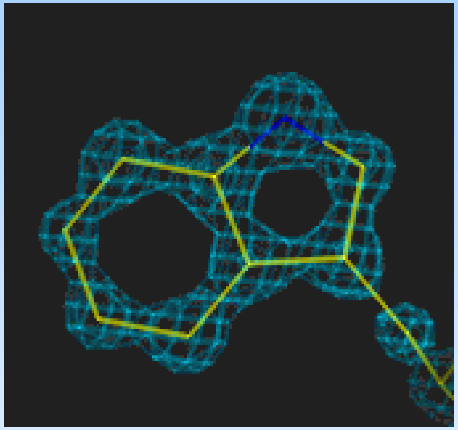
Here is the Fourier synthesis function:

$$f(x) = \frac{1}{\sqrt{2\pi}} \int_{-\infty}^{\infty} F(h) e^{-ihx}\, dh$$

When the limits of the summation are not so great, information is lost in the synthesized structure.

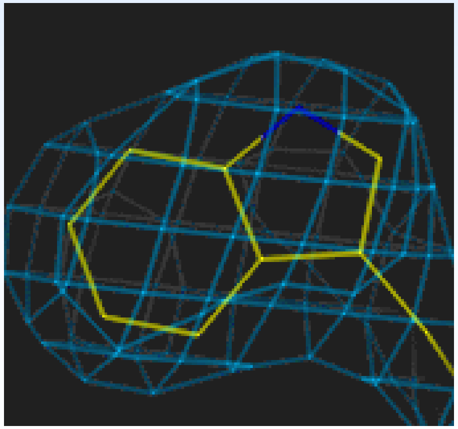
We say that the "**resolution**" equals the d-spacing of the smallest Bragg planes.

Resolution: The d-spacing of the highest order Bragg planes included in the Fourier synthesis. ***Small d-spacing is good.***

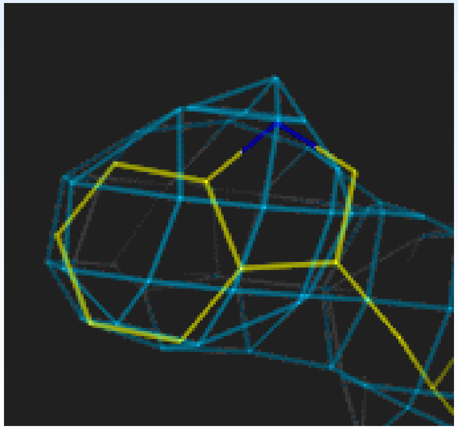


1.0 Ång (10,000 refl's)

1.8 Ång (1700 refl's)



3.0 Ång (370 refl's)



4.0 Ång (160 refl's)

Graphics by Phil Evans

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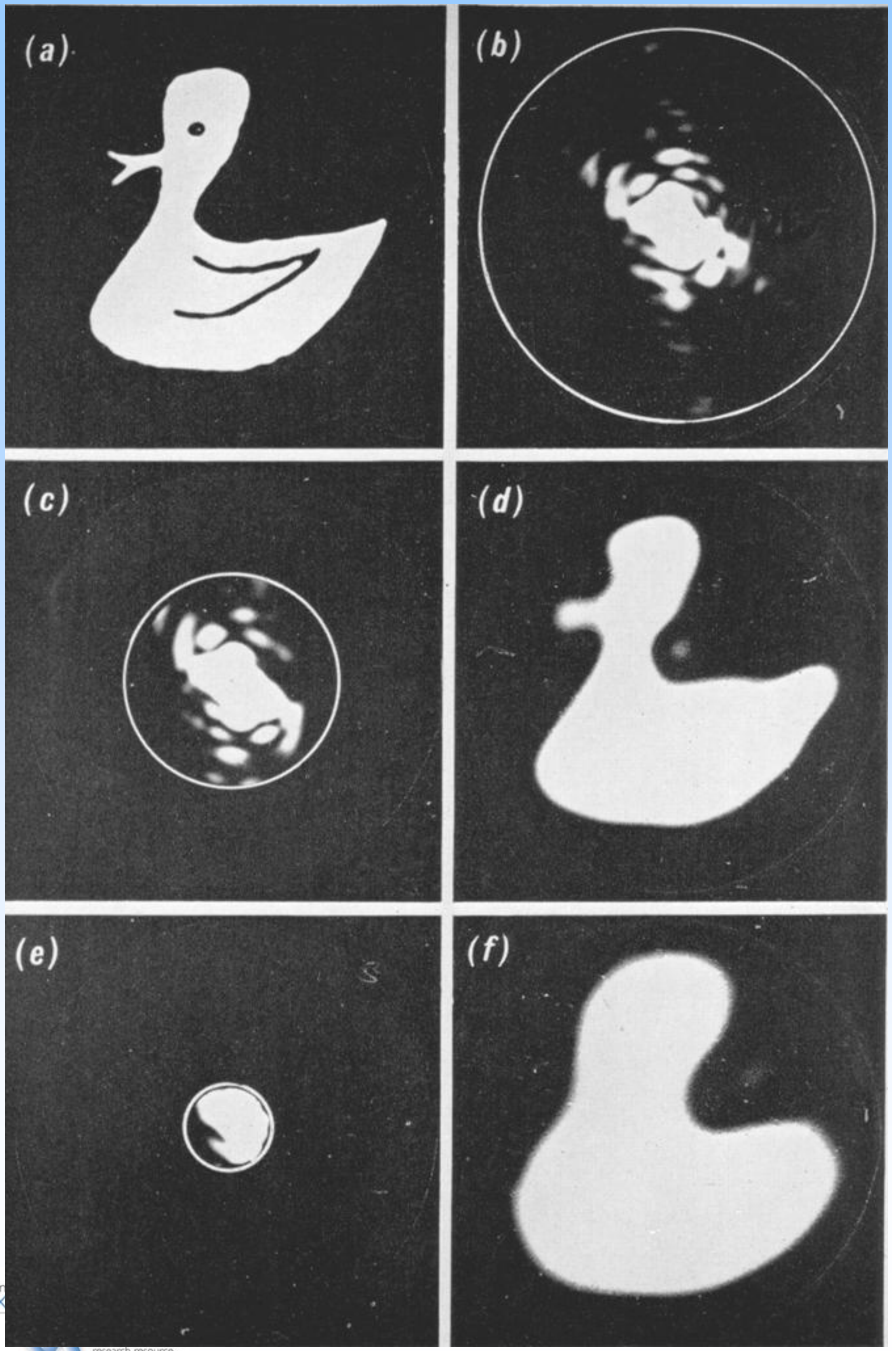


Another example.

The famous Taylor and Lipson rubber ducky.

Outline for the Lecture

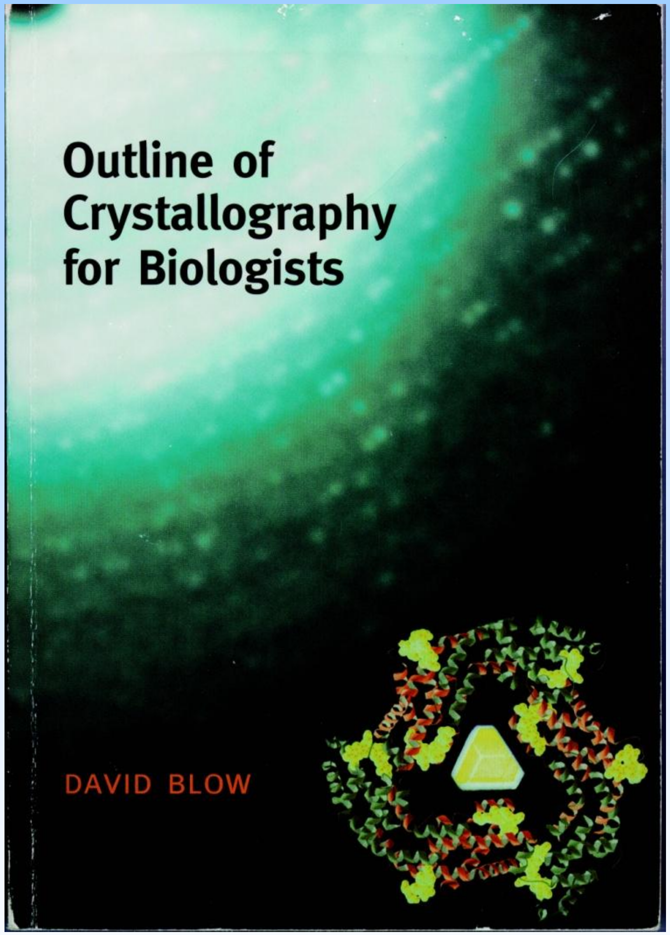
- Remind you how much you already know -- lenses, crystals
- Show why crystals give diffraction spots.
- Develop the idea of "The Reciprocal Lattice"
- Give some idea how we might actually measure diffraction data
- Show how, given a crystal, we can calculate the diffraction pattern
- Conversely, show how to calculate the structure from the diffraction
- **Describe the importance of symmetry to diffraction**
- Outline the structure-solving methods -- heavy atoms and MADness

Symmetry of crystals

We'll take some of our examples from David Blow's book.

Symmetry: **An operation of rotation, translation, inversion, mirroring, or some combination of these that takes an object back into itself.**

- The simplest symmetry in a crystal is repetition.
- The repeated motif may have its own symmetry.



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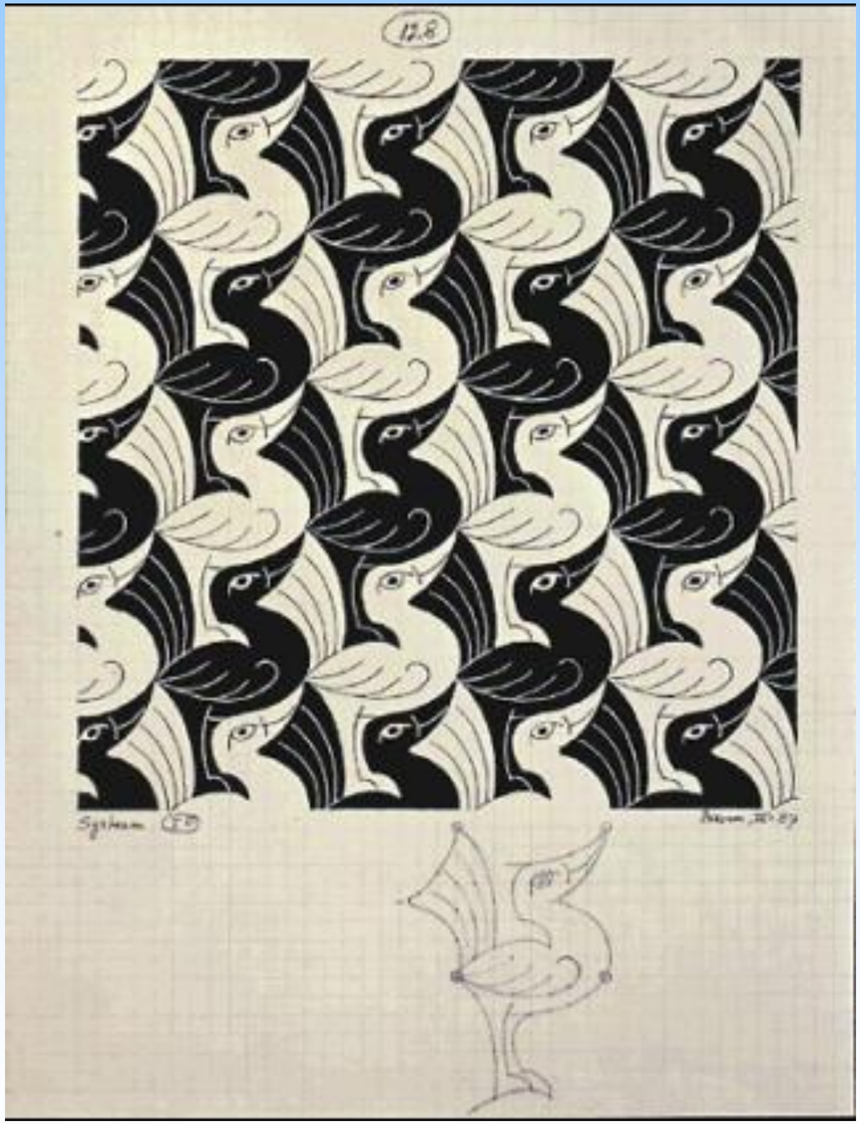
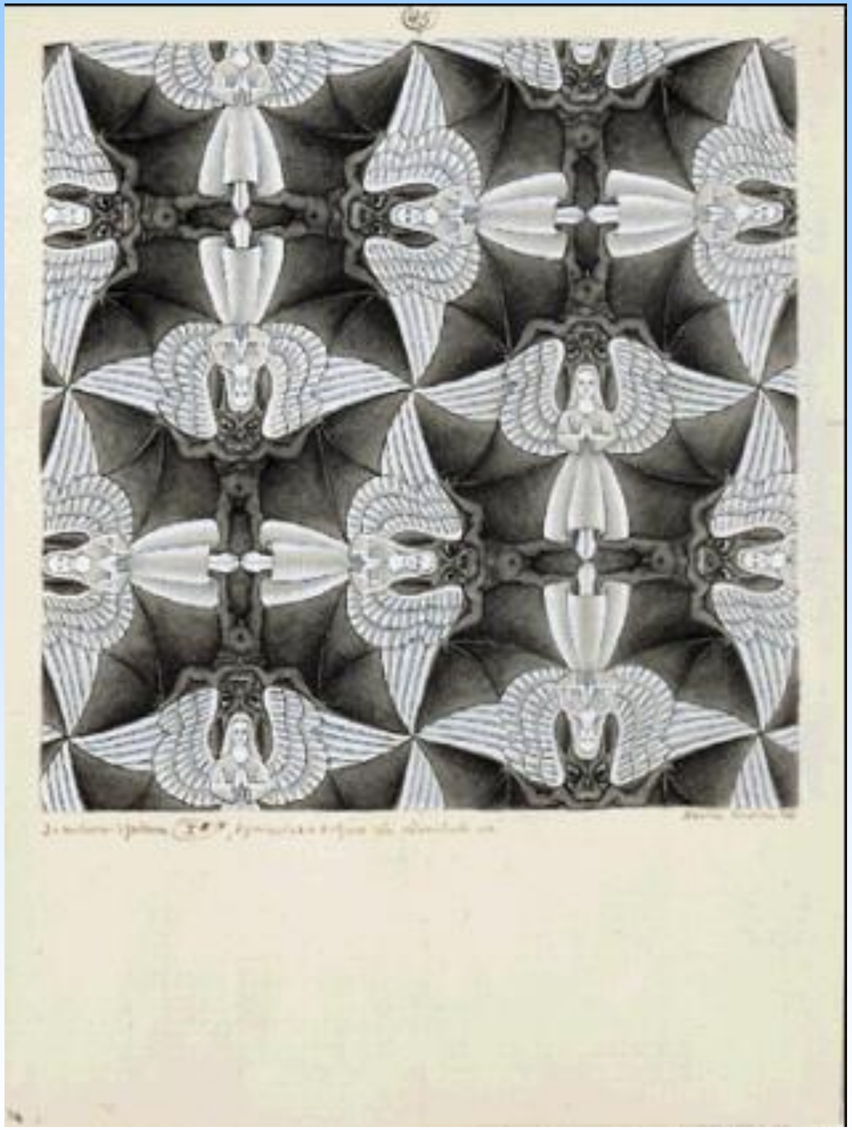
You know symmetry when you see it!

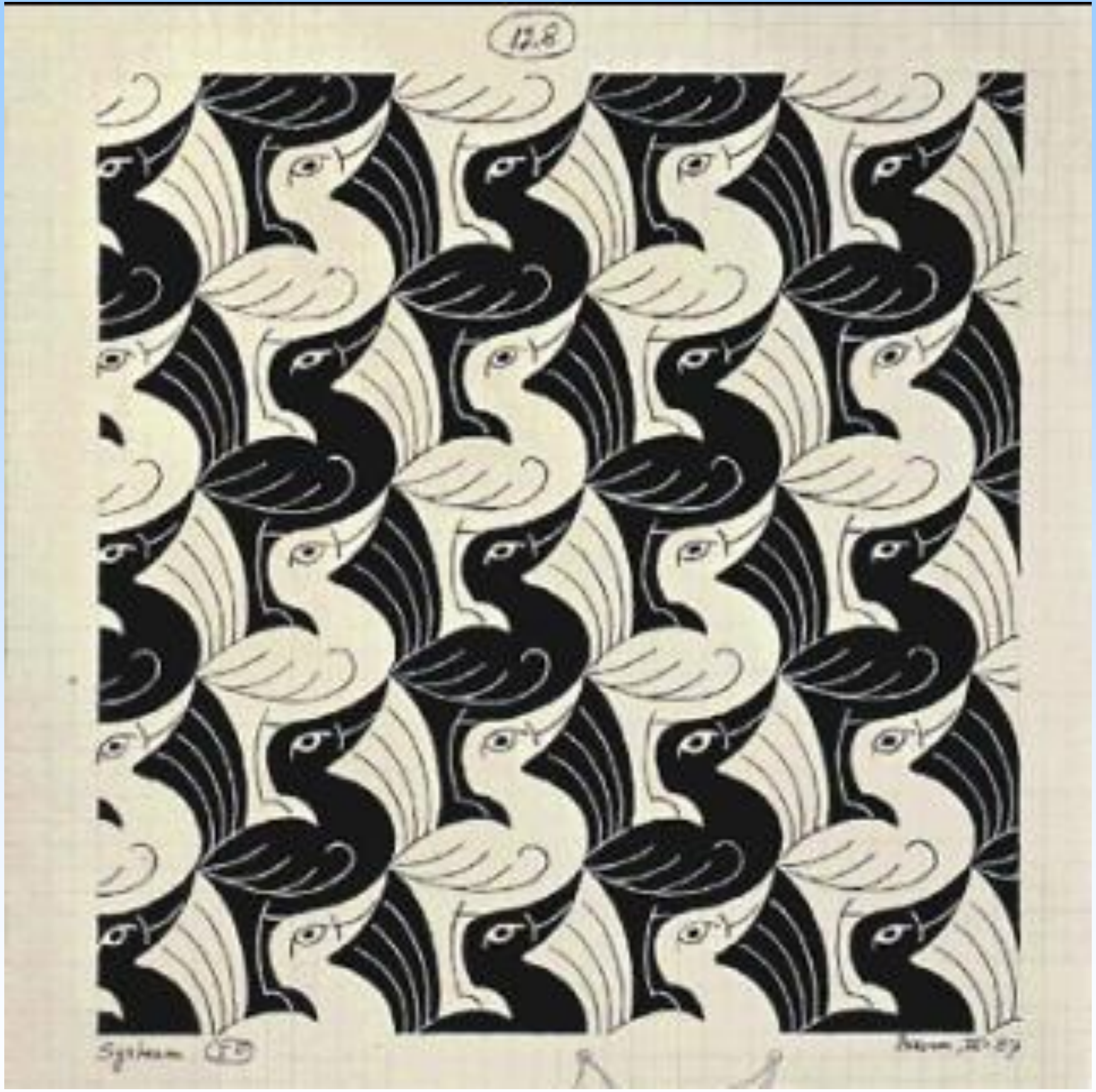




GRAPHICS BY M.C. ESCHER

http://www.mcescher.com/Gallery/gallery-symmetry.htm

Can we identify symmetry elements?



Graphics by M.C. Escher

What about here?

GRAPHICS BY M.C. ESCHER

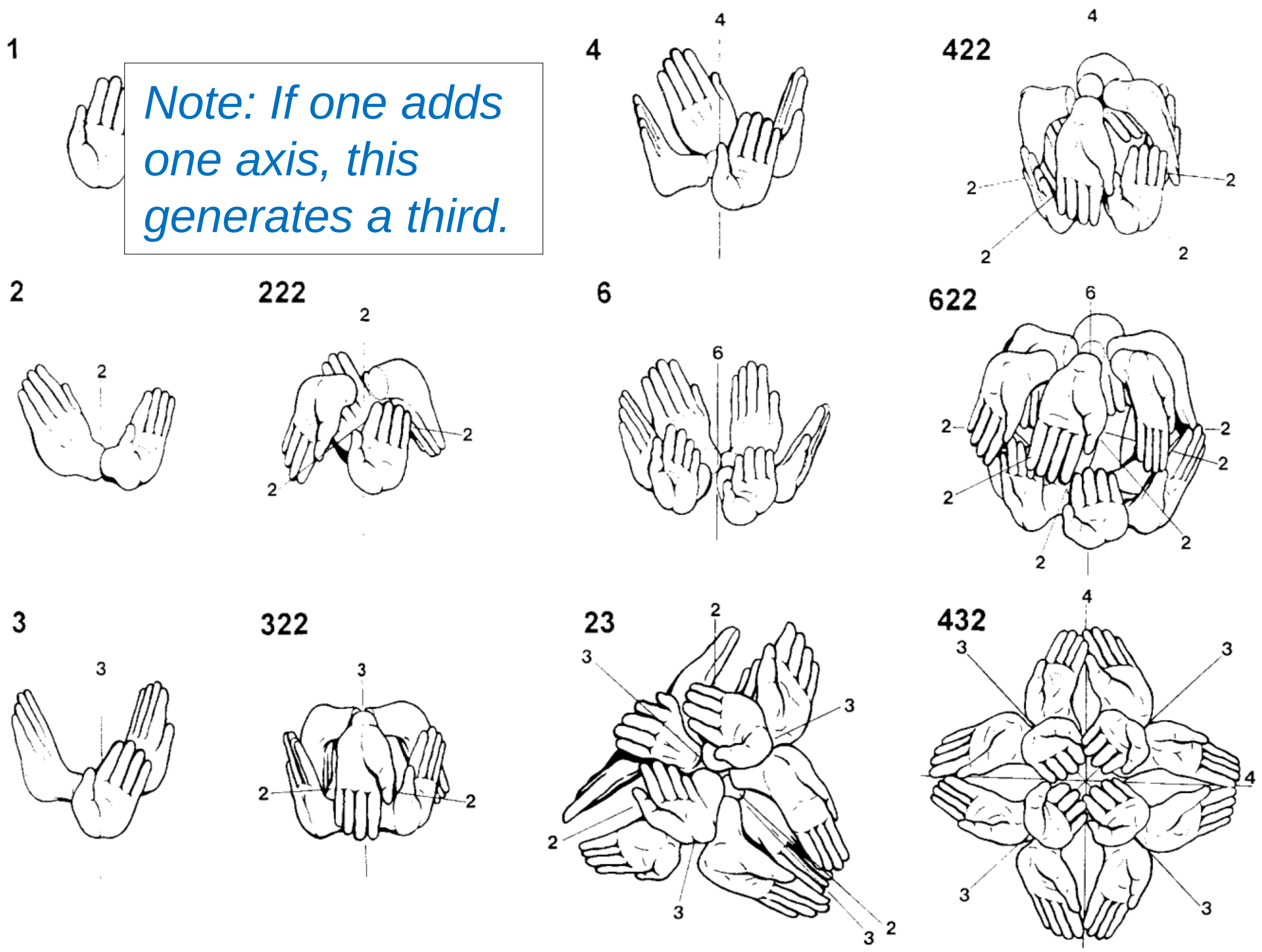
And here?

GRAPHICS BY M.C. ESCHER

And here?

GRAPHICS BY M.C. ESCHER

Symmetry Groups



Note: If one adds one axis, this generates a third.

Fig. 2.18 The point groups that can exist in protein crystals.

Biological molecules are all **chiral**, or “handed,” so **only rotation and translation symmetry are permissible**.

Here are the combinations (groups) of symmetries one finds in macromolecular crystals.

What is a Group?

Elements in a group must obey certain properties:

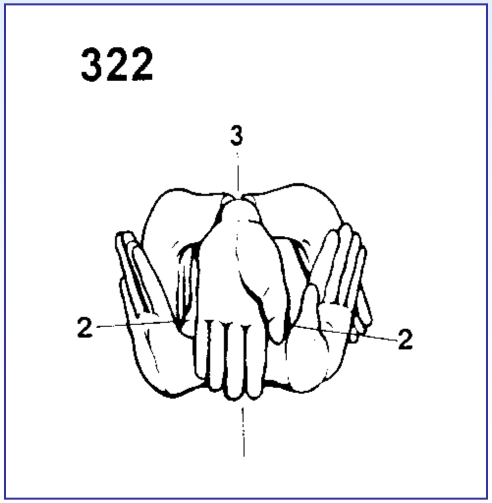
- There must be the ***identity*** element.
- The combination of any two elements must generate an element of the group. This is called ***closure***.
- Number of elements = number of objects repeated = ***order*** of the group.
- Every element in the group must have an ***inverse***.

$$3 \times 3^2 = I$$

$$3^2 = 3^{-1}$$

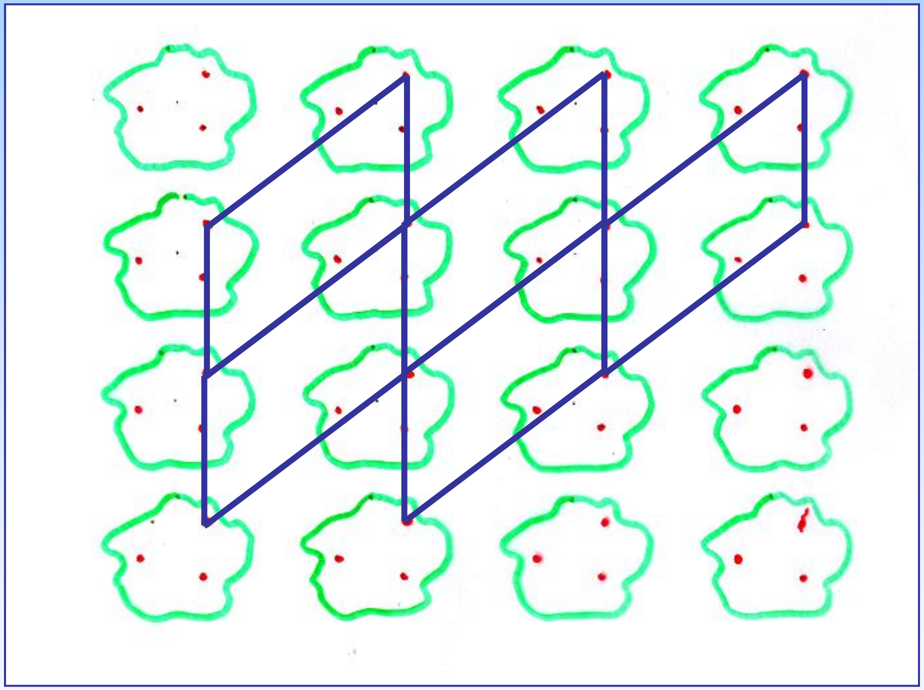
$$3 \times 2 = 2'$$

Point Group is **32**



Simple crystal symmetry

The simplest crystal would contain a single asymmetric object repeated by translational repetition only, like our apple orchard.



More complicated crystal symmetry

A crystal could contain a symmetric object, also repeated by translational repetition.

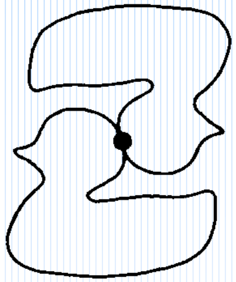


Fig. 2.35 A symmetrical dimer.

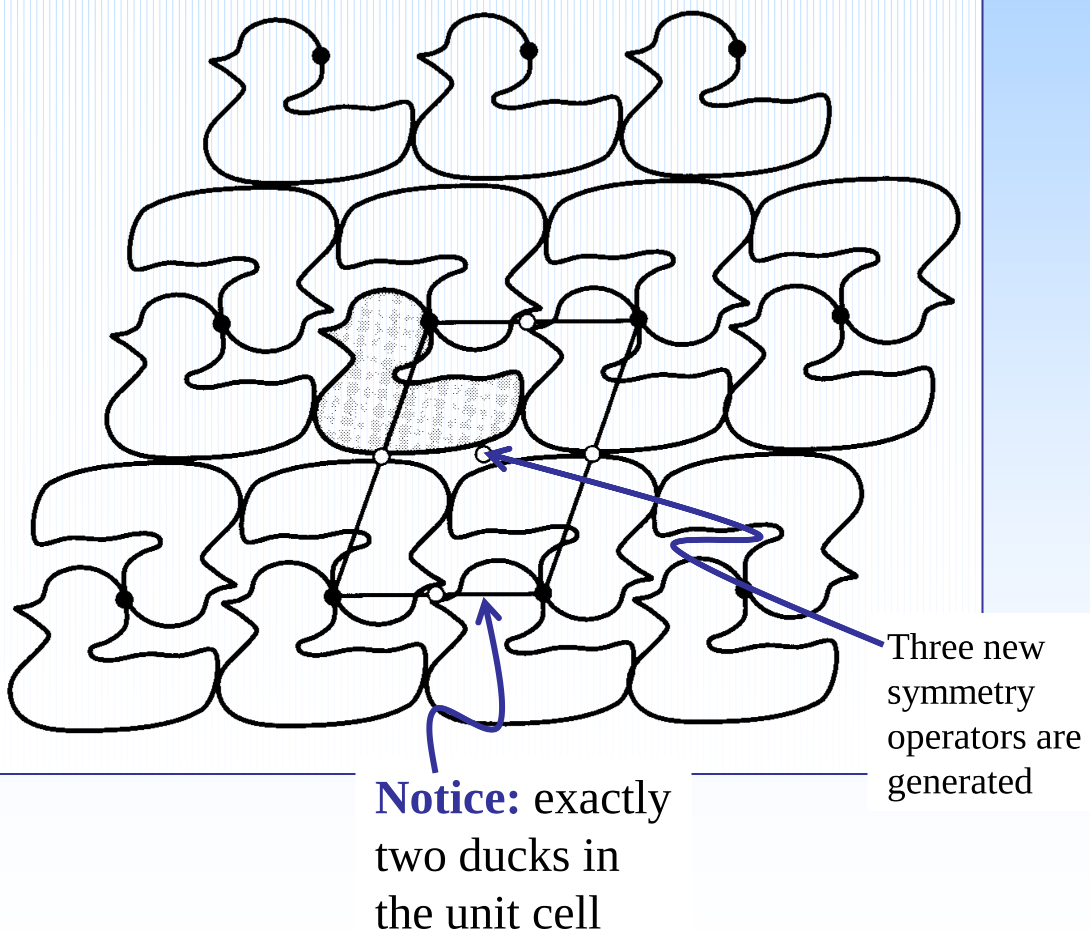


Fig. 2.36 The smallest unit of the structure that can generate the complete crystal structure by means of its symmetry operations is called the crystal asymmetric unit.

Now let's try it in three dimensions

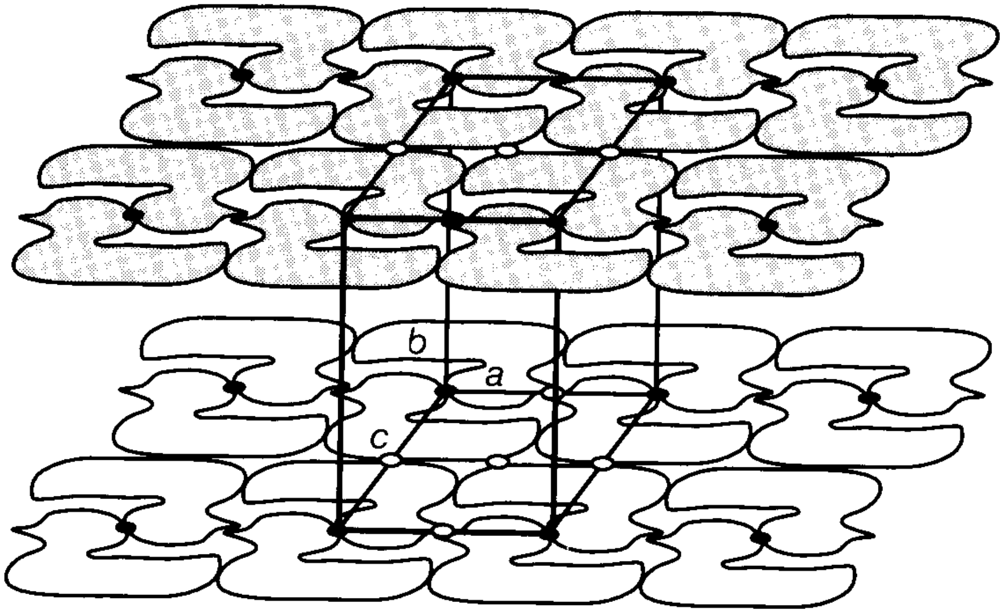


Fig. 2.37 Symmetry and equivalent positions in space group P2. A 2-fold axis along ***b*** creates two asymmetric units in the unit cell. Each unit has four 2-fold axes associated with it, at $x,z = (0, 0)$ (black circles), and at (0, 1/2), (1/2, 0), (1/2, 1/2) (open circles).

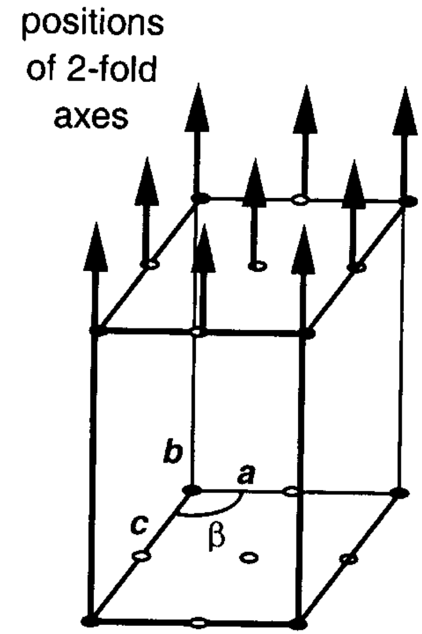


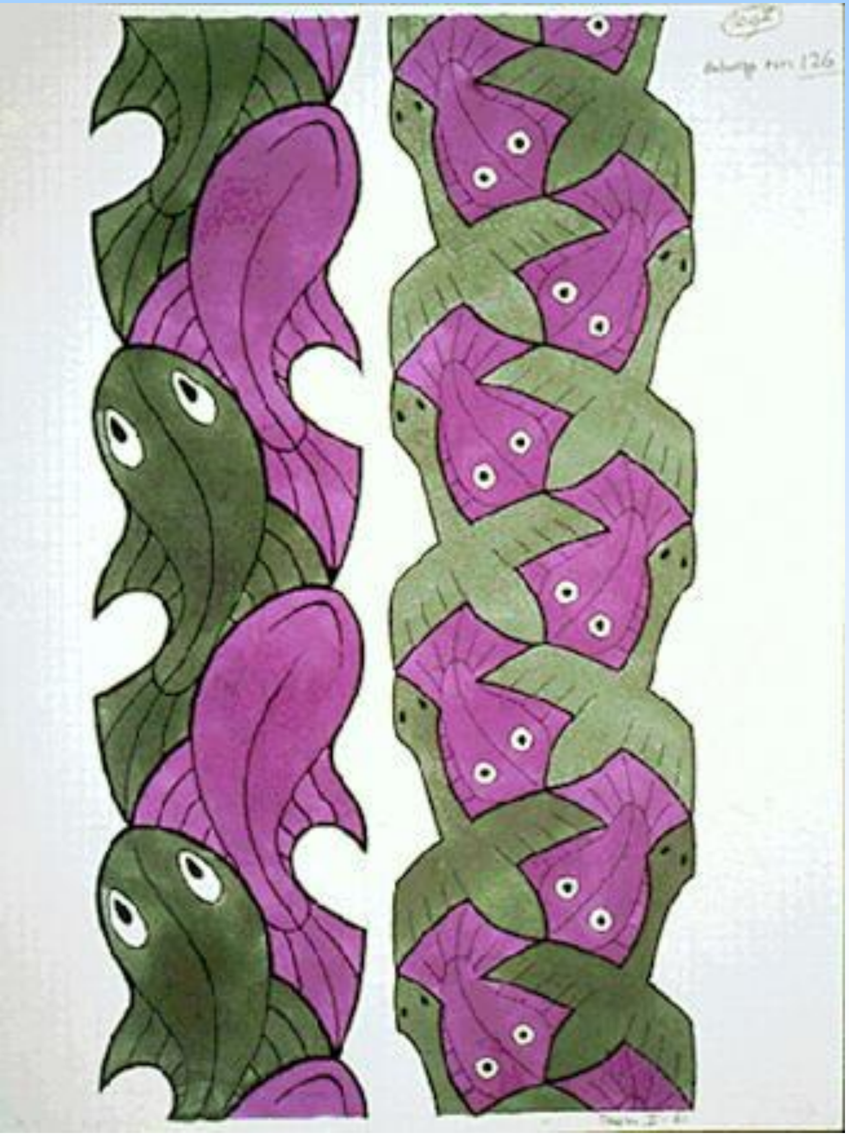
Fig. 2.38 A unit cell of space group P2.

Space Group P2: **P** = "primitive," **2** = two-fold rotation axis.

We call this type of crystal ***monoclinic***. ***Order*** = 2.

Can we create an operation that combines two simple operations into a compound one?

Rotation then translation is a ***screw axis.***



Mirroring then translation is a ***glide plane.***

The Screw Axis

This symmetry operation is an **m**–fold rotation followed by a translation.

The translation is a **n/m** translation along one of the major crystallographic directions, where **m** is the order of the major rotation axis: the $\mathbf{m_n}$ screw axis.

Here, it's written $\mathbf{2_1}$ to represent the two-fold screw axis, and the translation is ½.

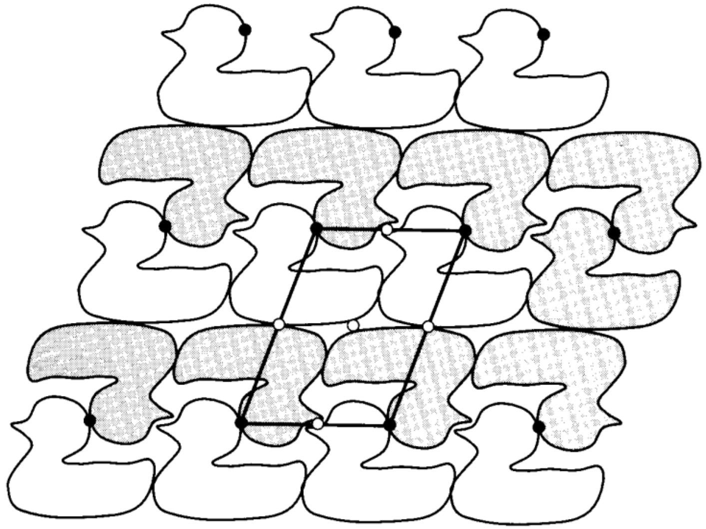


Fig. 2.43 A $P2_1$ structure viewed down the ***b*** direction. The unshaded molecules are at $y = 0$, and the shaded molecules at $y = 1/2$. There are 2-fold screw axes at the corners of the unit cell, and also at positions indicated by white circles.

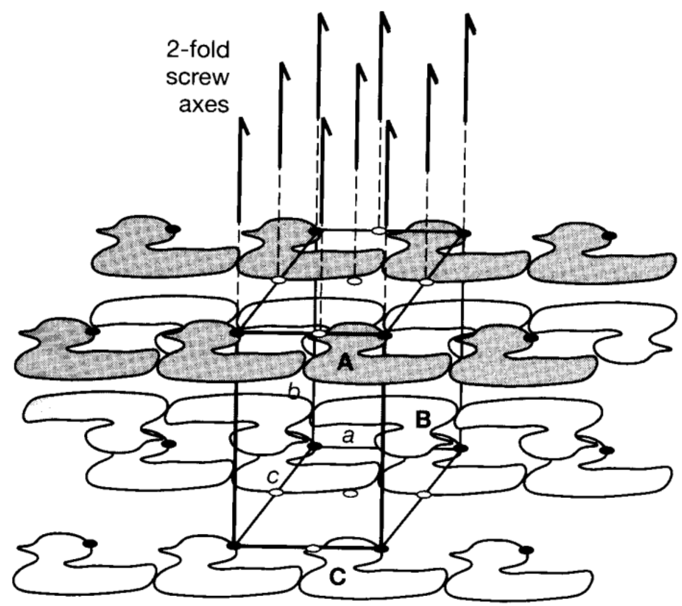


Fig. 2.44 Arrangement of units in a $P2_1$ lattice. Units facing one way are at the top and bottom of the cell, those facing the other are halfway in between. Objects A, B, and C are related by a 2-fold screw operation.

Centered Lattice

To make a new ***monoclinic*** lattice, shift the motif at the origin along a diagonal to a new spot by a major fraction of the unit cell edges.

The lattice is “centered” because a new motif appears in the center of a face or of the body of the unit cell.

When P2 is “centered” to form C2, new **2_1** axes are formed.

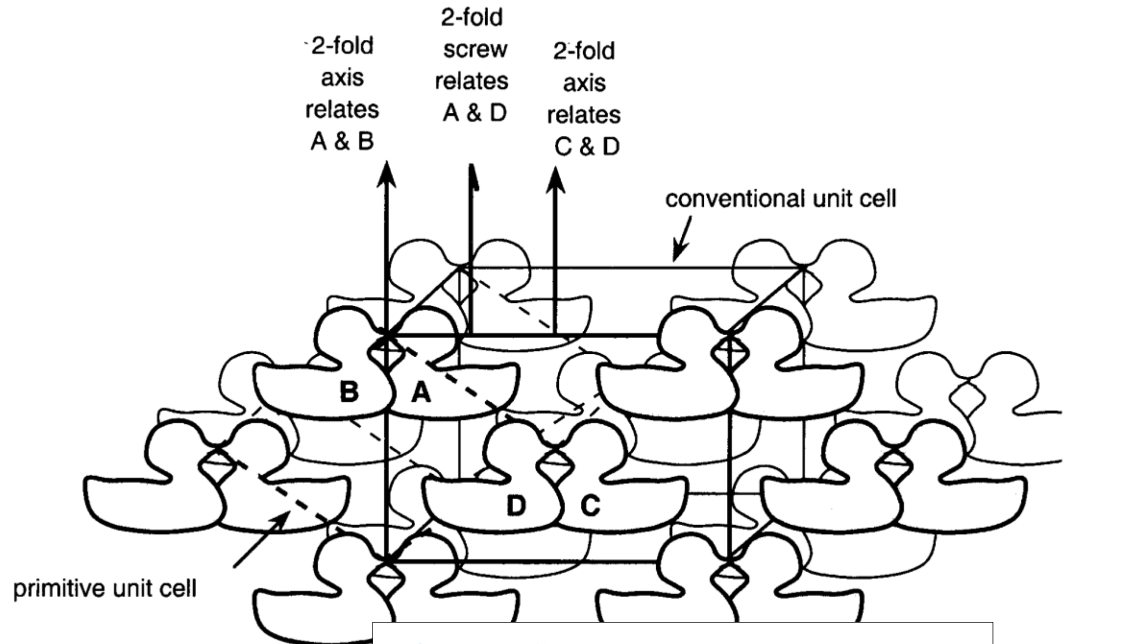


Fig. 2.39 A C-face centred monocli

*If one “centers” the **B** face, one simply generates a new **P2** unit cell.*

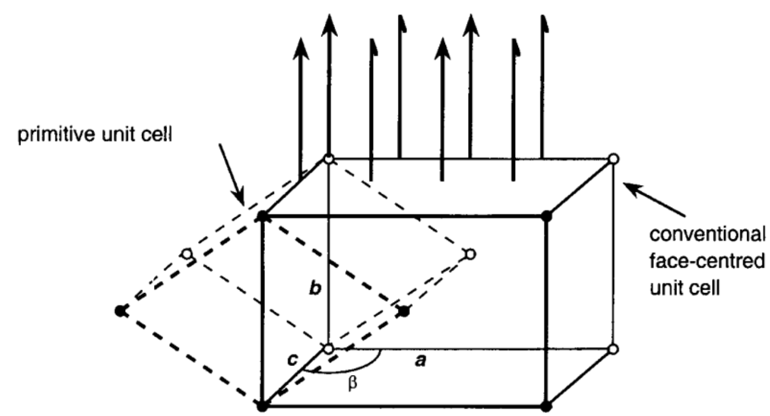


Fig. 2.40 Summary of the symmetry of a C2 lattice. Screw axes are identified by single-barbed arrows.

And higher symmetry

If one has two-fold axes in more than one direction, it must be three directions, and the axes must be perpendicular. We call this ***orthorhombic***.

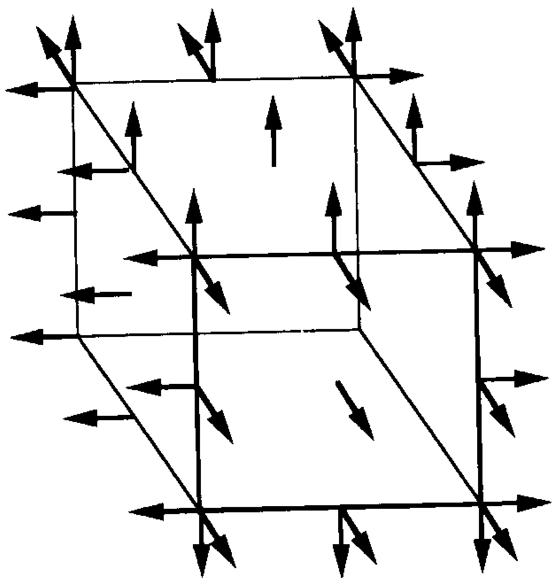


Fig. 2.41 P222 has intersecting sets of 2-fold axes in three perpendicular directions.

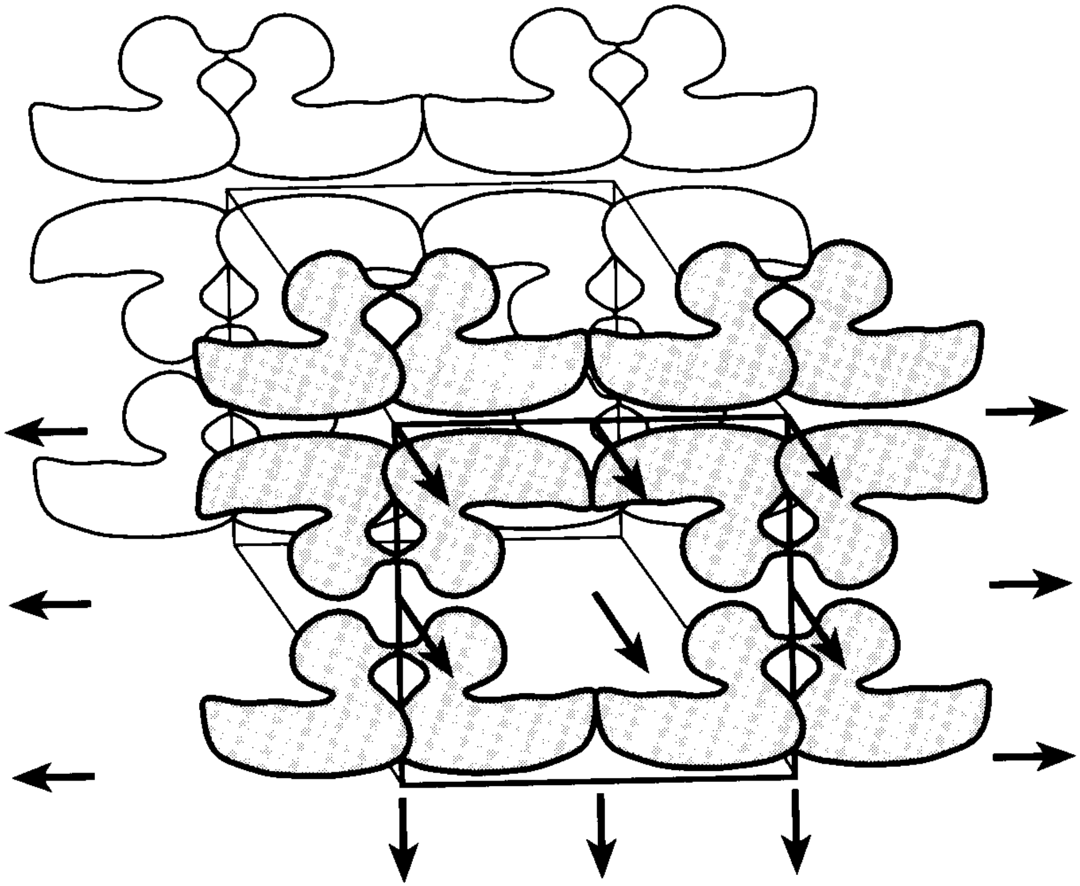


Fig. 2.42 Molecular arrangement in space group P222, showing just a few of the 2-fold axes.

And finally …

A three-fold axis will produce a ***trigonal*** crystal. Notice how the first three-fold axis creates two other three-folds with different environments.

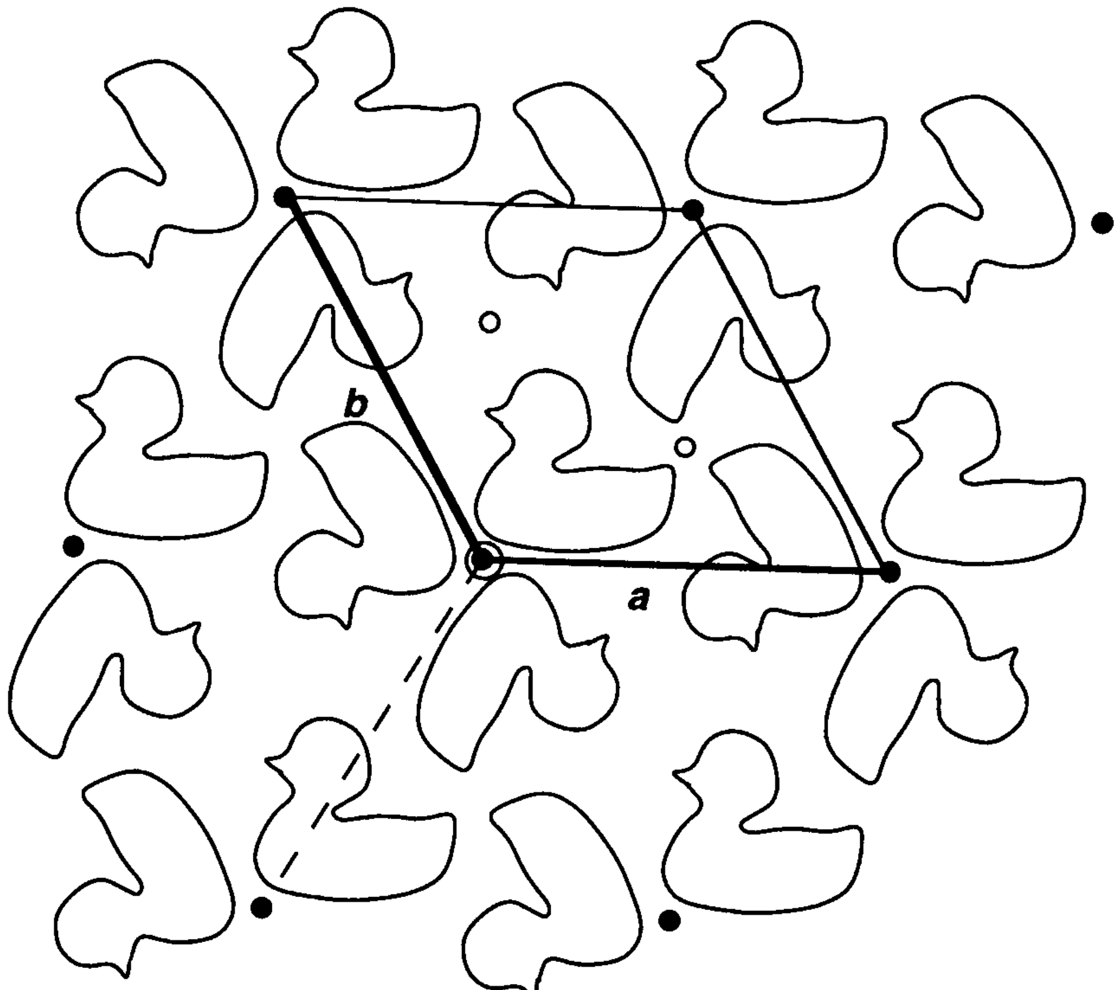


Fig. 2.28 If there is 3-fold symmetry, the lattice is generated by two lattice translations which make an angle of 120° and are of equal length. When objects are arranged with 3-fold symmetry about the lattice points, two other types of 3-fold symmetry axis are generated, indicated within the outlined cell.

The Seven Crystal Systems

The combination of symmetry elements yields only these forms

Crystal System	Bravais Types	External Minimum Symmetry	Unit Cell Properties
Triclinic	P	None	a, b, c, al, be, ga,
Monoclinic	P, C	One 2-fold axis, parallel b (b unique)	a, b, c, 90, be, 90
Orthorhombic	P, I, F, C	Three perpendicular 2-folds	a, b, c, 90, 90, 90
Tetragonal	P, I	One 4-fold axis, parallel c	a, a, c, 90, 90, 90
Trigonal	P*, R	One 3-fold axis	a, a, c, 90, 90, 120
Hexagonal	P*	One 6-fold axis	a, a, c, 90, 90, 120
Cubic	P, F, I	Four 3-folds along space diagonal	a, a, ,a, 90, 90, 90

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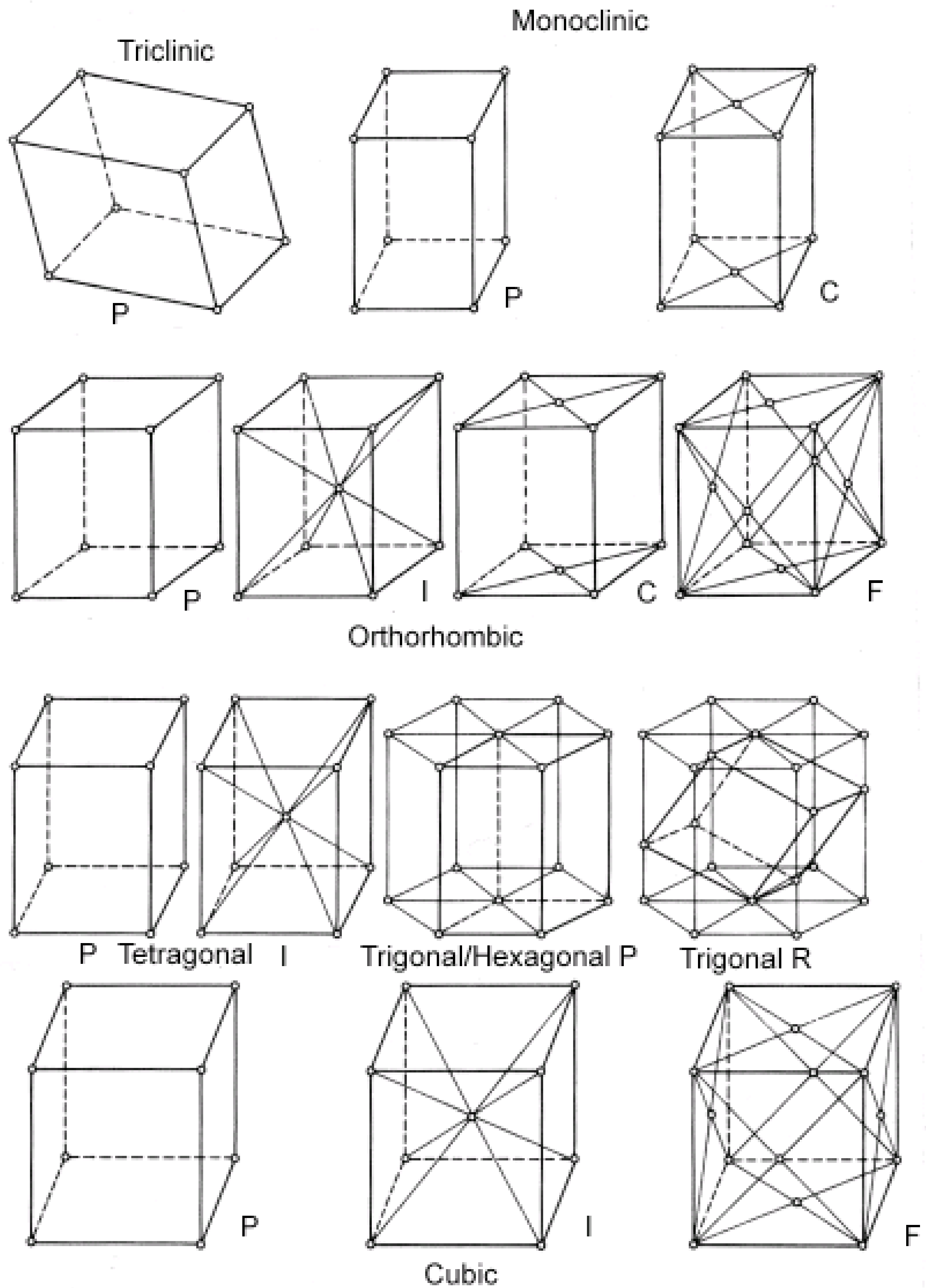
** Note: P(hexagonal) = P(trigonal)*

The Bravais Lattices

Here are the 14 ways crystal lattices can be formed in the seven crystal systems.

The international convention in displaying these is to give **a** down, **b** across, and **c** up or towards the viewer.



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How many space groups?

- There are 230 space groups possible
- Only 65 of these employ only rotational symmetry **(suitable for chiral molecules)**
- Here are the most abundant observed in macromolecular structures, 65% of the total:

Space group symbol	% of total
$P2_12_12_1$	24.2
$P3_221$ & $P3_121$	15.2
$P2_1$	13.8
C2	6.1
$P4_32_12$	5.4

And finally the icosahedral symmetry of spherical viruses

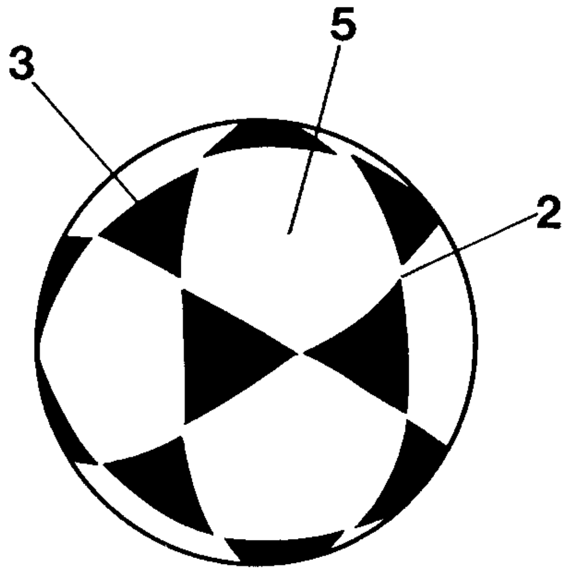

Fig. 2.19 Footballs are often decorated in a way that shows 532 symmetry.

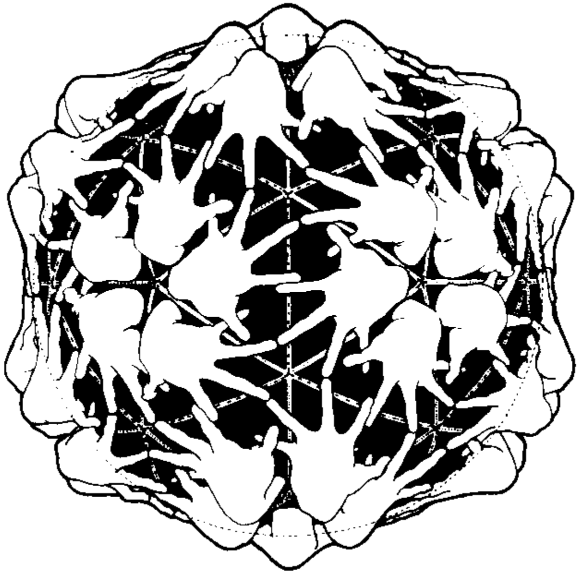

Fig. 2.20 Fanciful drawing of left hands arranged in 532 symmetry by Don Caspar (reproduced from Caspar (1980) by permission of the Biophysical Society).

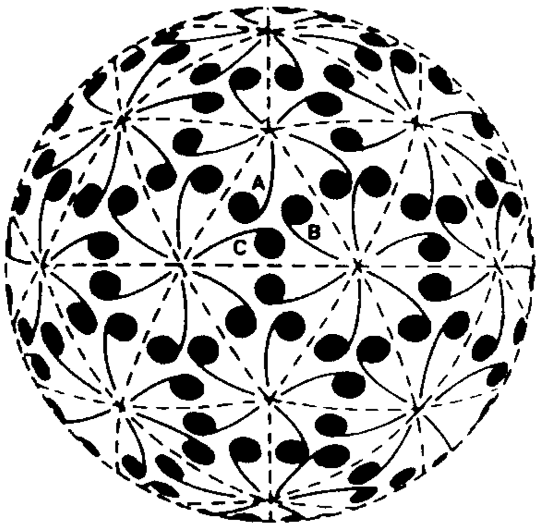

Fig. 2.21 Pseudo-symmetrical arrangement of 180 units (reproduced from Harrison (1980) by permission of the Biophysical Society).

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How does symmetry affect a diffraction pattern?

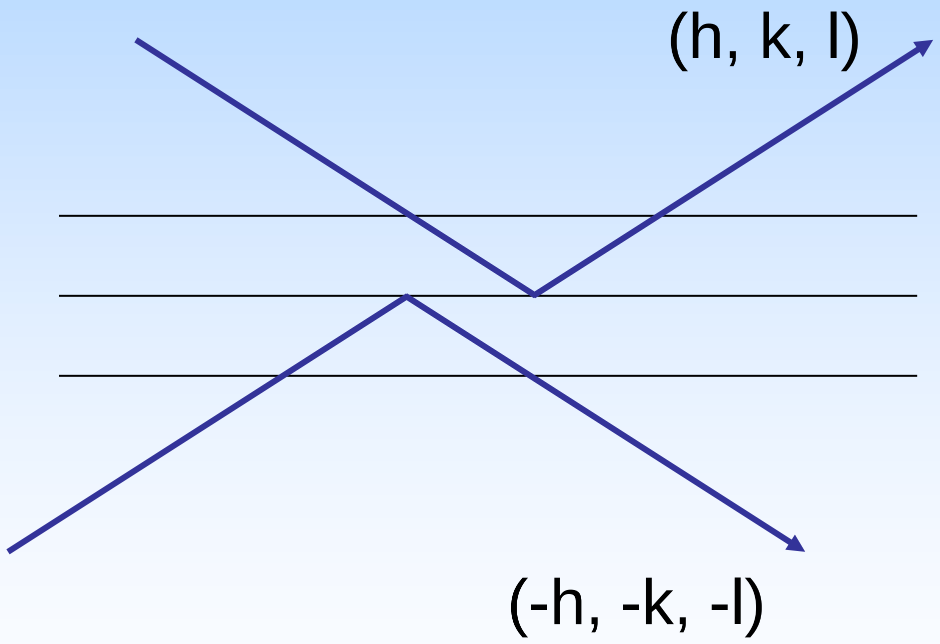
Symmetry affects a diffraction pattern in at least three ways:

Friedel's Law – There's an inversion centre in reciprocal space.

Laue Point Group – Diffraction has symmetry like that of the crystal.

Systematic absences – some of the symmetry operations erase some reflections.

Friedel's Law: Bragg reflection from the front of the planes is the same as from the back.



We can do this algebraically

Friedel's Law: Intensity is the same for (hkl) and $(\bar{h}\bar{k}\bar{l})$

$$F_{\bar{h}\bar{k}\bar{l}} = \sum_{\text{atoms}} f_j \exp\left[2\pi i(-hx_j - ky_j - lz_j)\right]$$

$$= \sum f_j \exp(-2\pi i \, \underline{h} \cdot \underline{r}_j)$$

$$= \sum f_j \left[\cos(-2\pi \, \underline{h} \cdot \underline{r}_j) + i \cdot \sin(-2\pi \, \underline{h} \cdot \underline{r}_j)\right]$$

$$= \sum f_j \left[\cos(2\pi \, \underline{h} \cdot \underline{r}_j) - i \cdot \sin(2\pi \, \underline{h} \cdot \underline{r}_j)\right]$$

$$= F^*_{hkl}$$

Do you know abt. complex conjugates?

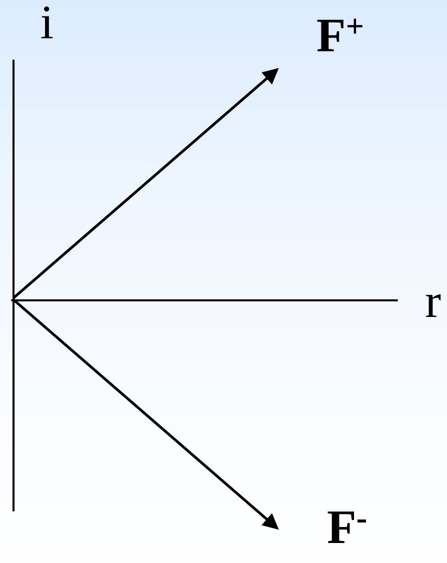
For $F = A + iB$

$F^* = A - iB$

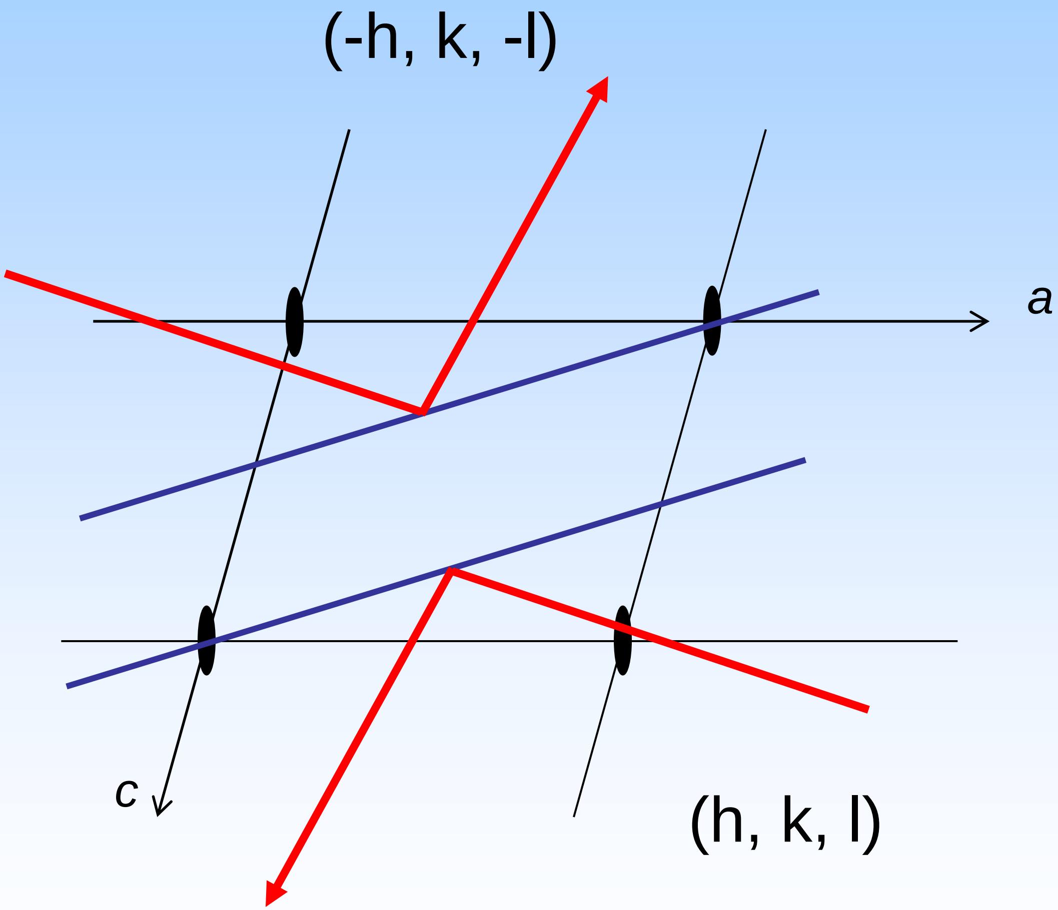
and $|F| = (F \cdot F^*)^{1/2} = (A^2 + B^2)^{1/2}$

Therefore $|F_{hkl}| = |F_{\bar{h}\bar{k}\bar{l}}|$:

Friedel's Law



Laue Point Group: The diffraction will adopt some of the symmetry of the crystal.



Let's say the (1, 5, 2) and the (-1, 5 –2)

The **Laue Point Group** for a crystal is the rotational or mirror symmetry of the space group, plus Friedel's Law. For example:

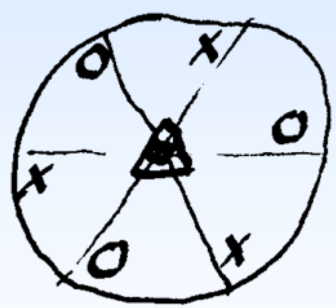
$$\mathbf{P2} \text{ or } \mathbf{P2_1} \rightarrow \mathbf{2/m}$$

Produces a two-fold, a mirror perpendicular to it, and an inversion centre ***in the diffraction pattern / reciprocal space***.

How to describe symmetry – Stereographic Projection

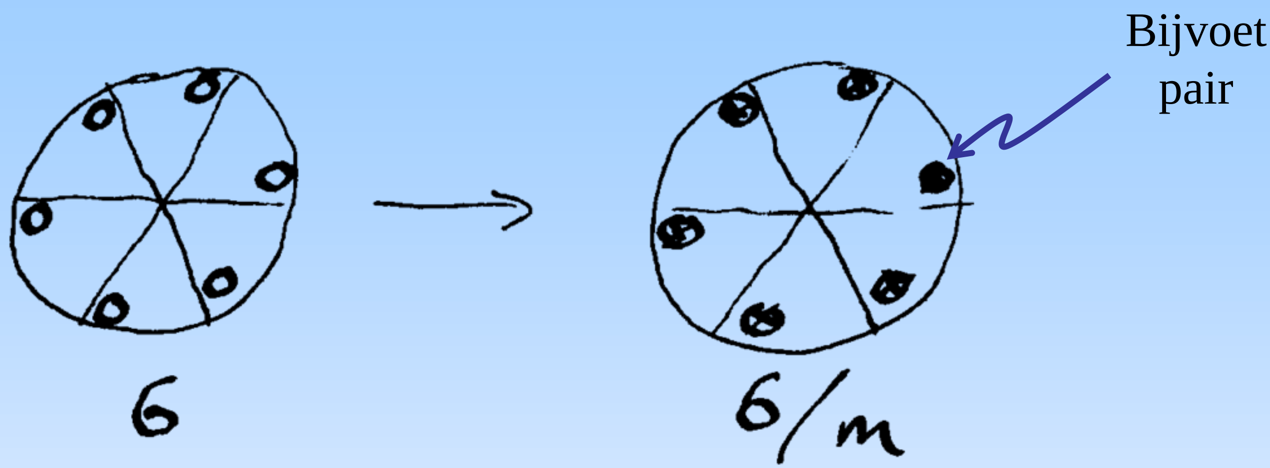
$2/m$

$\bar{3}$



3-fold, followed by inversion

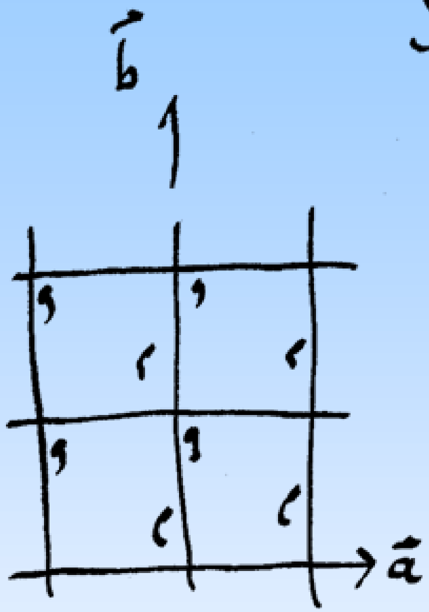
Laue point group for $P6_1$



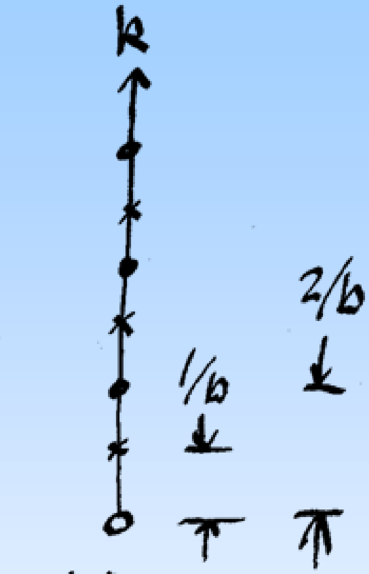
Implication of this for the experiment:

One will need only to record 1/12 of reciprocal space to get complete data. Sometimes one can record anomalously-related reflections on the same image.

Systematic Absenses



Compress onto b axis

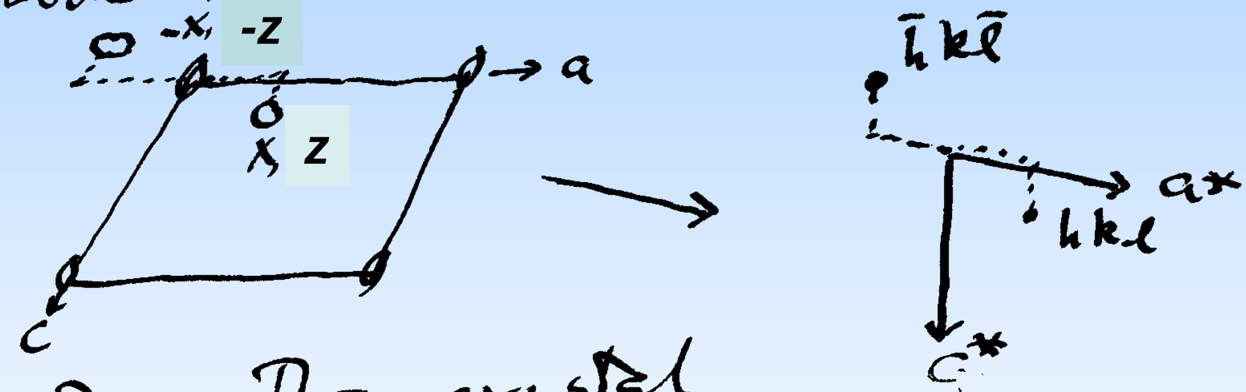


0k0 ray in the diffn pattern

We say, for 0k0,
$k = 2n$

We also can try to understand how symmetry operations affect the symmetry of the diffraction pattern.

Look at P2:



Does this crystal symmetry produce the diffraction symmetry we predict?

Evaluate the structure factor for P2:

$$F_{hkl} = \sum_{n/2} f_j \Big[\cos 2\pi(hx_j + ky_j + lz_j) + i\cdot\sin 2\pi(hx_j + ky_j + lz_j)$$
$$+ \cos 2\pi(-hx_j + ky_j - lz_j) + i\cdot\sin 2\pi(-hx_j + ky_j - lz_j)\Big]$$

Then, use: $\sin(x \pm y) = \sin x \cdot \cos y \pm \cos x \cdot \sin y$

and $\cos(x \pm y) = \cos x \cdot \cos y \mp \sin x \cdot \sin y$

To get:

$$F_{hkl} = 2\sum_{n/2} f_j \cdot \cos 2\pi(hx_j + lz_j)\cdot\left[\cos 2\pi k y_j + i\cdot\sin 2\pi k y_j\right]$$

One can see that $F_{hkl} = F_{\bar{h}k\bar{l}}$ as advertised

Also notice that for ***h 0 l*** data, there is NO imaginary part to the structure factor. The structure factor is **PURE REAL.**

Outline for the Lecture

- Remind you how much you already know -- lenses, crystals
- Show why crystals give diffraction spots.
- Develop the idea of "The Reciprocal Lattice"
- Give some idea how we might actually measure diffraction data
- Show how, given a crystal, we can calculate the diffraction pattern
- Conversely, show how to calculate the structure from the diffraction
- Describe the importance of symmetry to diffraction
- **Outline the structure-solving methods -- heavy atoms and MADness**

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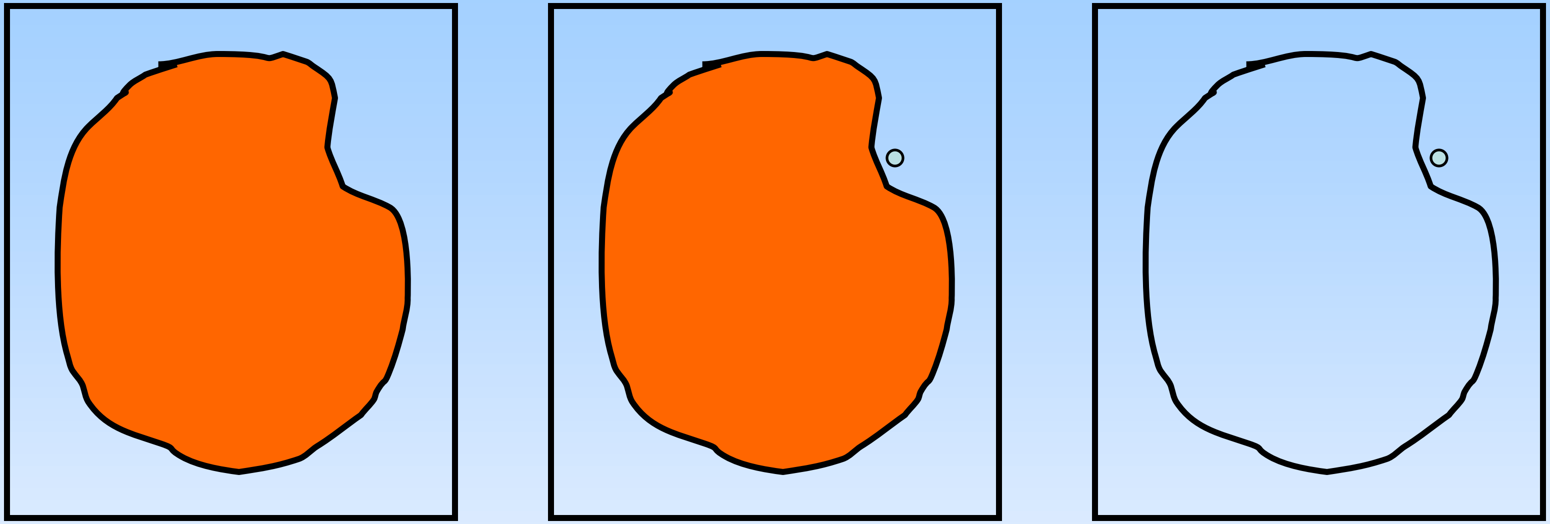
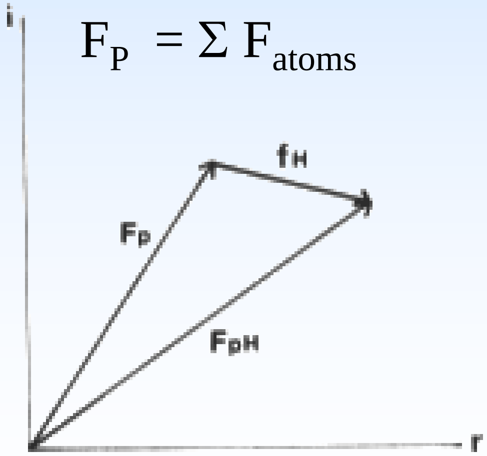
How we solve structures? We must somehow estimate phases so we can perform the inverse Fourier transform.

- Isomorphous Replacement with heavy atoms
- MAD/SAD, a variant of IR
- Molecular replacement if we have a decent model.

Perutz's Fundamental Idea:
Isomorphous Replacement

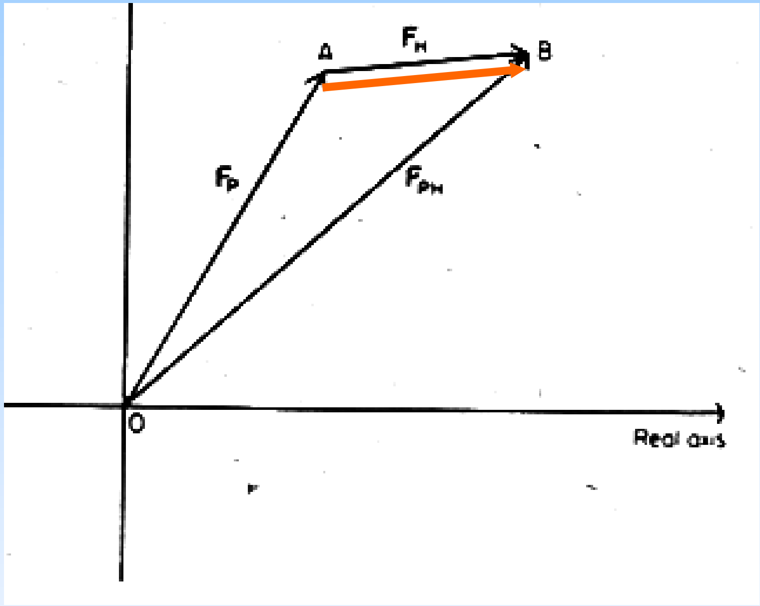


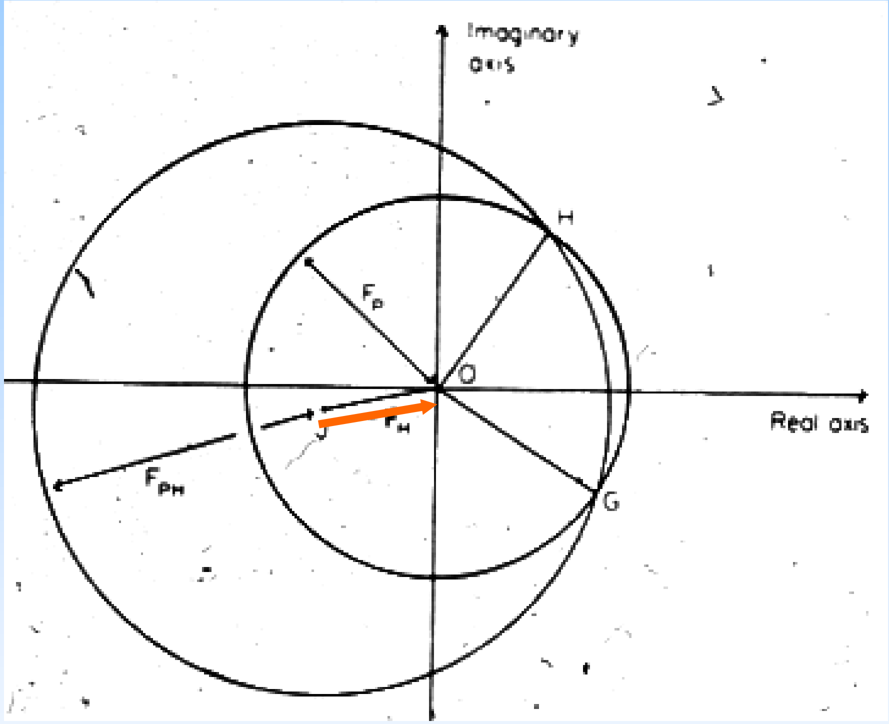
$F_P = \Sigma F_{atoms}$ $F_{PH} = F_P + F_H$ F_H



We find that, for some things, we can approximate $|F_H|$ with $|F_{PH} - F_P|$. This often suffices for us to solve for the positions of the heavy atom as if it were a small-molecule structure.

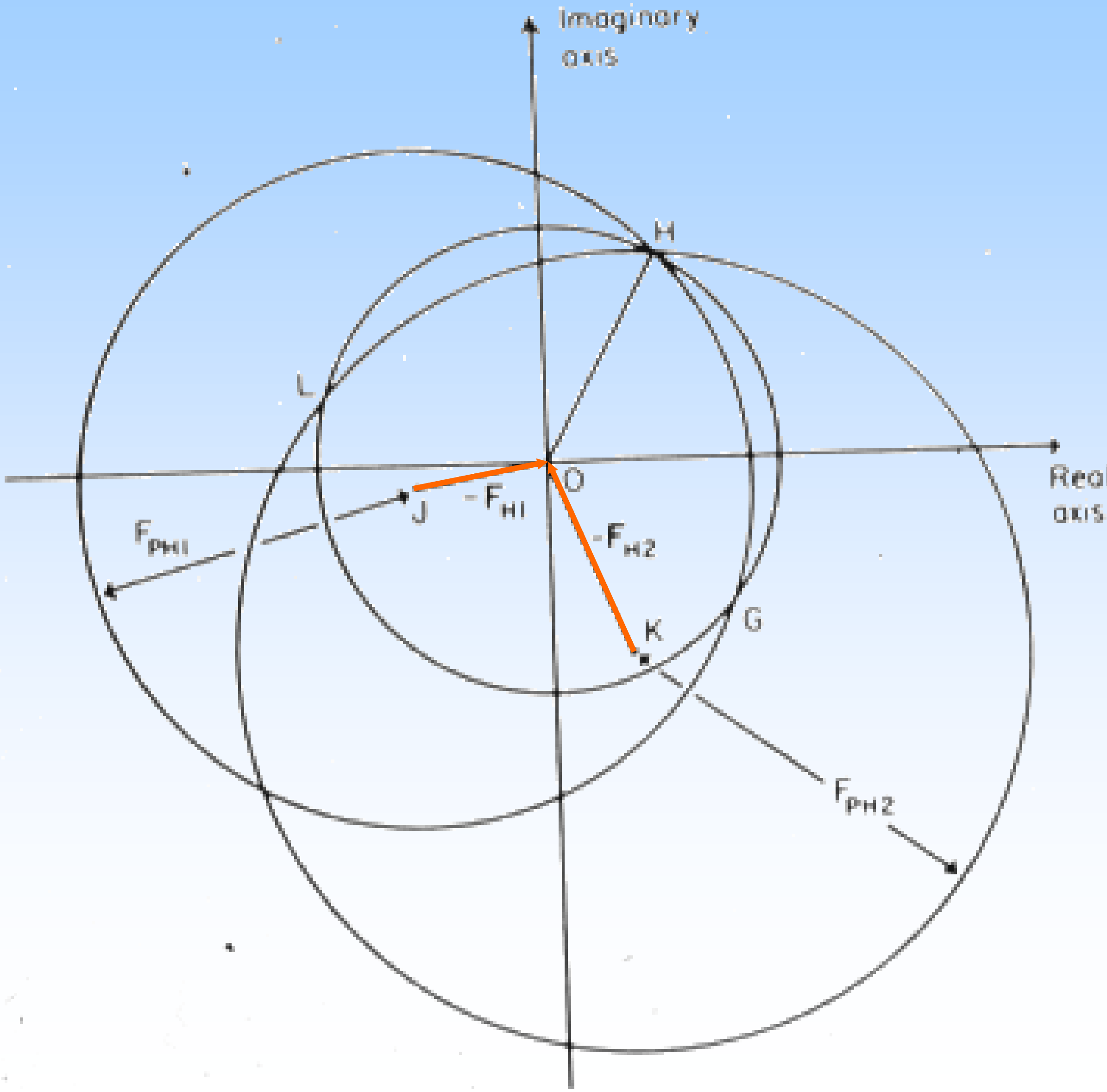
So for some particular reflection and a particular heavy atom, we can begin to find the phase:





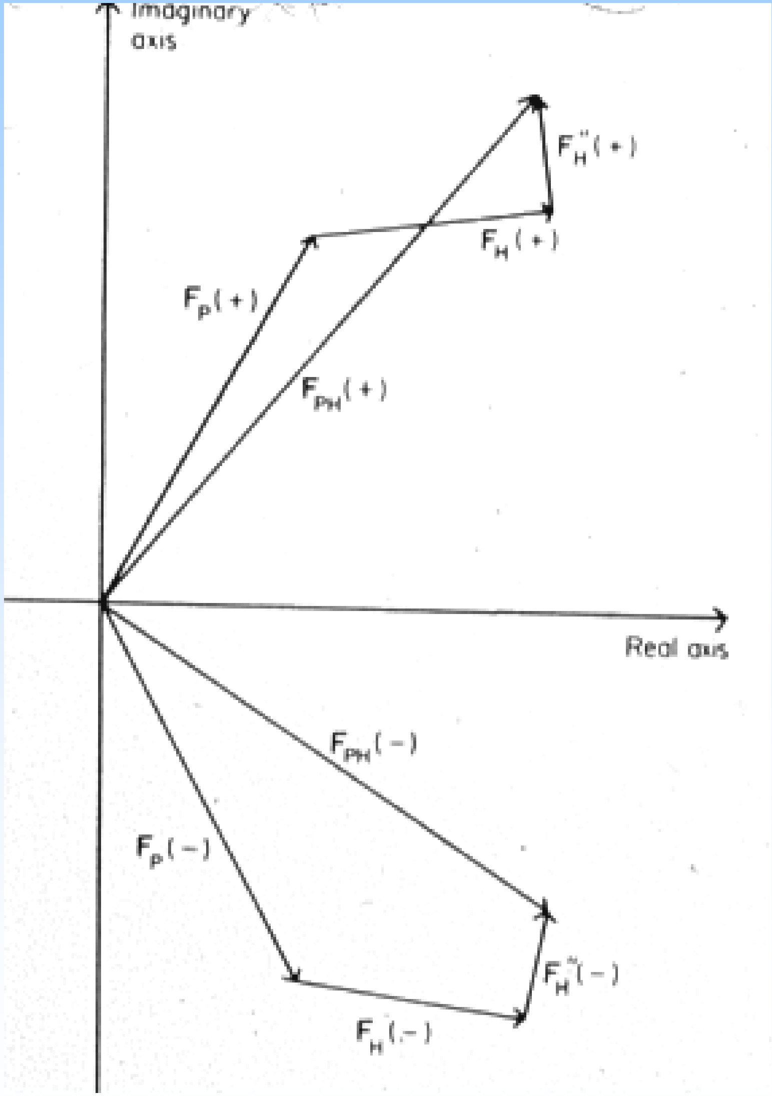
Knowing the position of the heavy atom allows us to calculate F_H. Then we use $F_P = F_{PH} + (-)F_H$ to show that the phase triangles close with a **two-fold ambiguity**, at G and at H. There are several ways to resolve the ambiguity.

One way to resolve the ambiguity is to use a second isomorphous heavy-atom derivative.



A second technique involves use of anomalous (resonant) scattering from a heavy atom.

In this case the resonance between the electrons on the heavy atom and the x-rays cause a phase and amplitude shift. The symmetry of diffraction (from the front *vs* back of the Bragg planes) is broken. **Friedel's Law is broken!** This can be measured and used.



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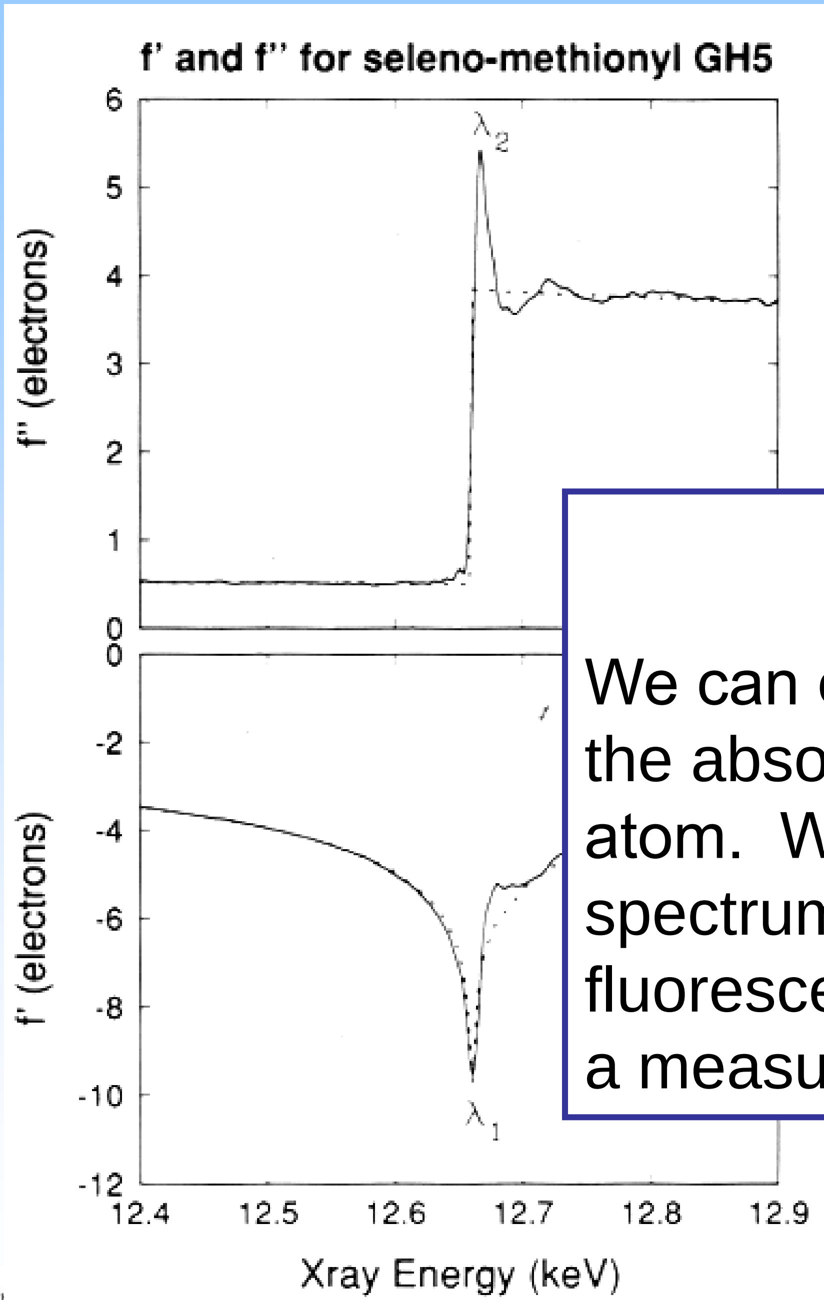
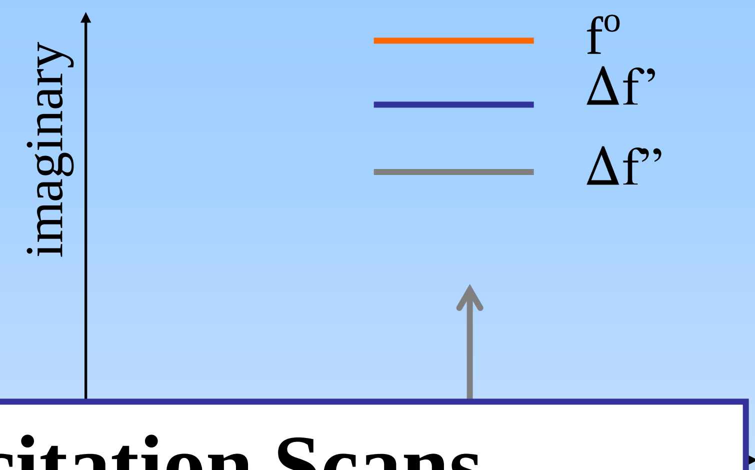


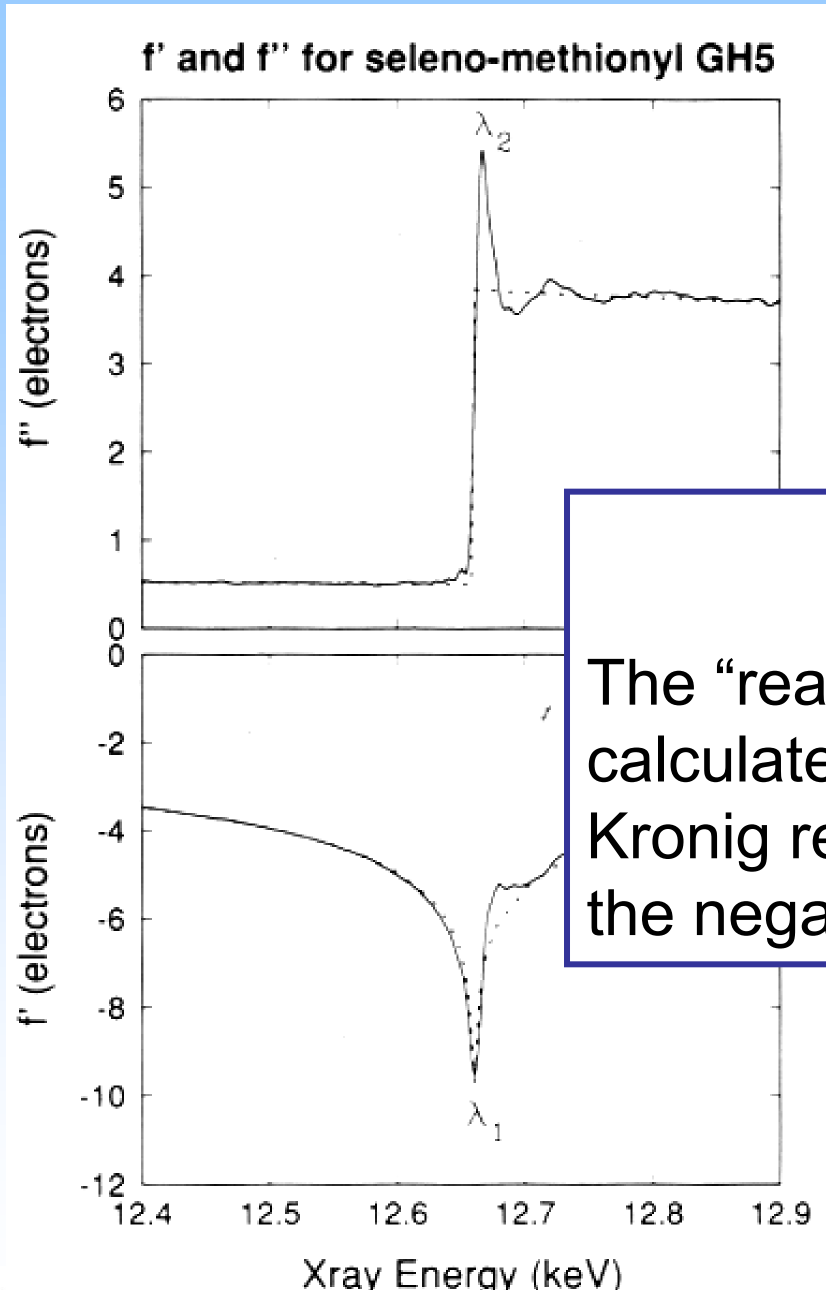
From Ramakrishnan's study of GH5



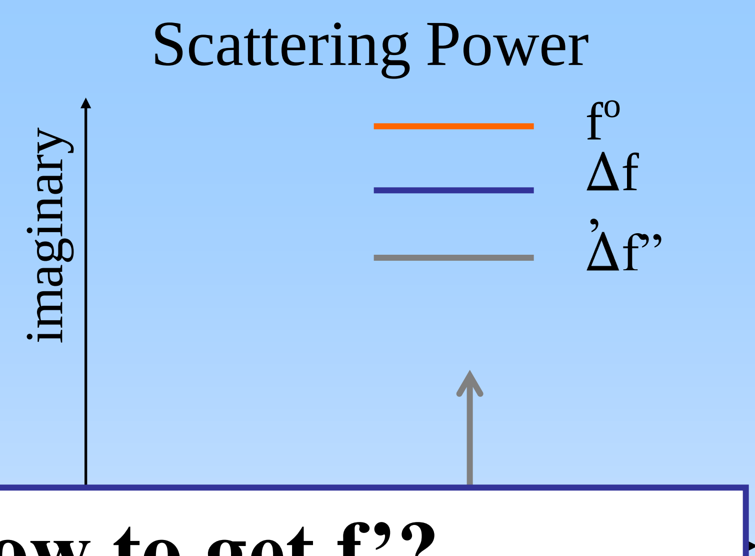
Excitation Scans

We can observe the f" by measuring the absorption of the x-rays by the atom. We measure an "excitation" spectrum. Often we us the fluorescence of the absorbing atom as a measure of absorptivity.

x-rays as a function of the photon energy.



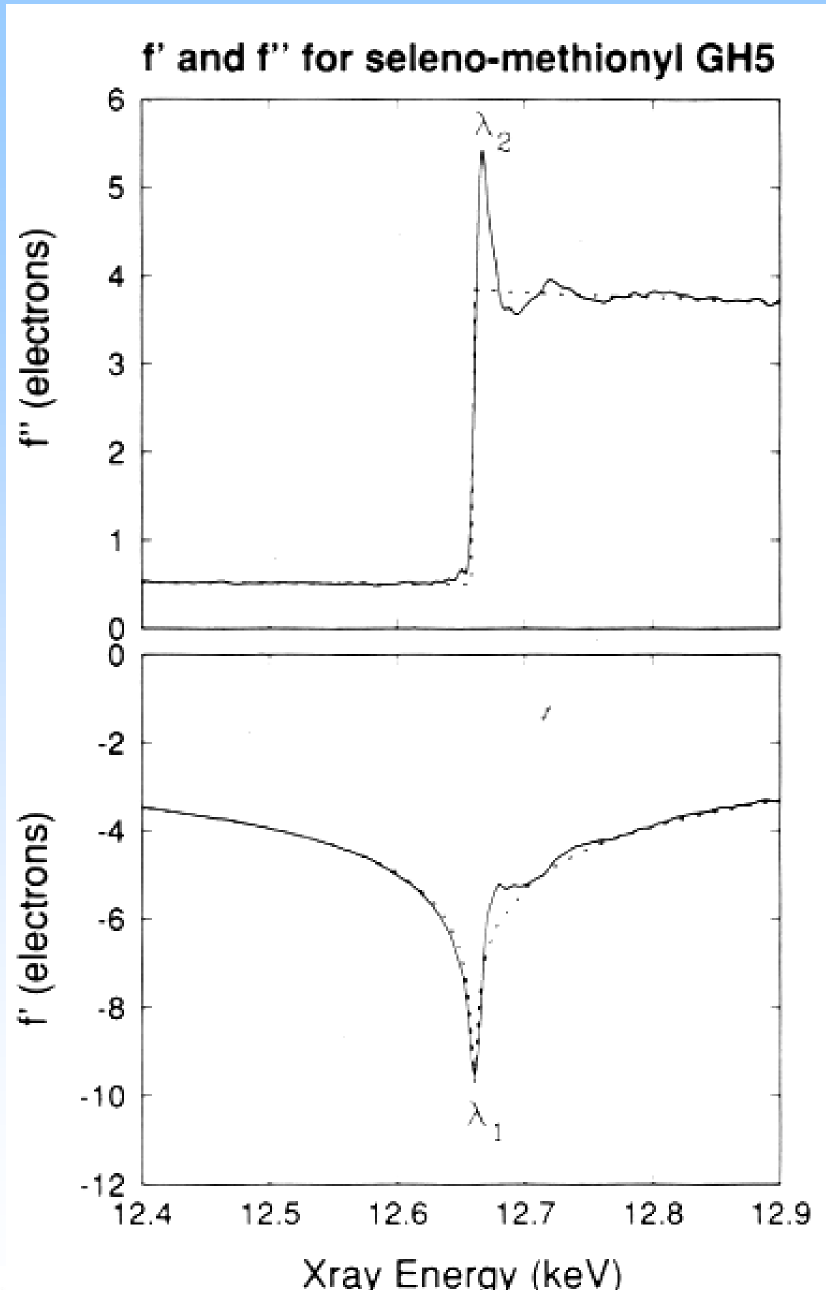
From Ramakrishnan's study of GH5



How to get f'?

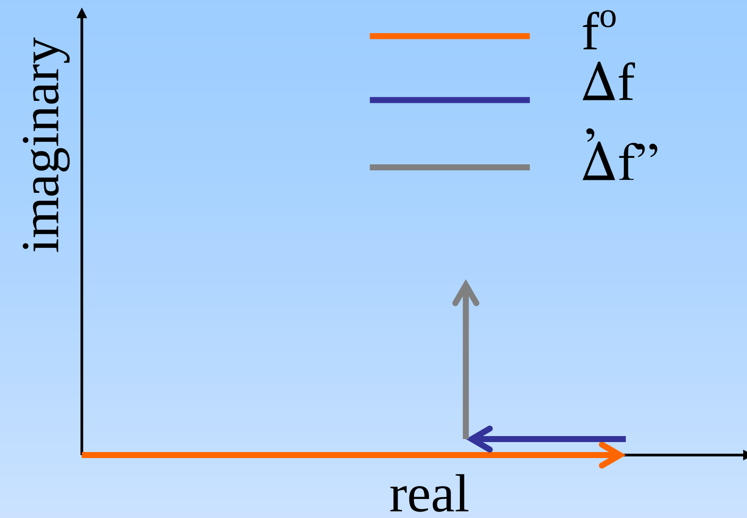
The "real," dispersive component is calculated from f" by the Kramers-Kronig relationship. Very roughly, it's the negative first derivative of f".

(Δf'') of the scattering of x-rays as a function of the photon energy.



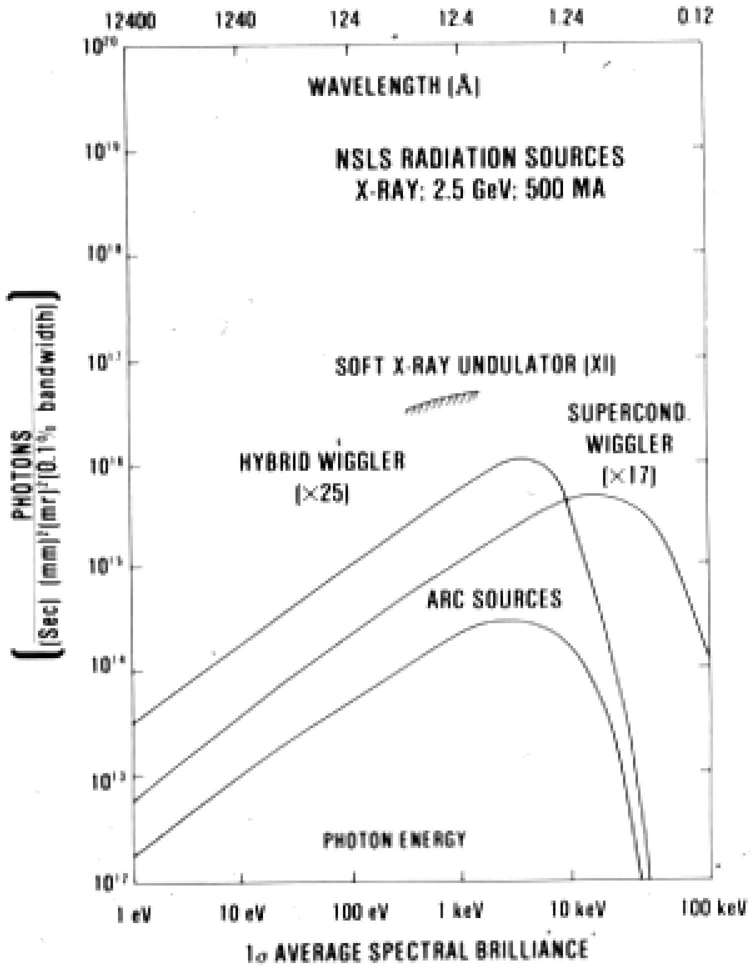
From Ramakrishnan’s study of GH5

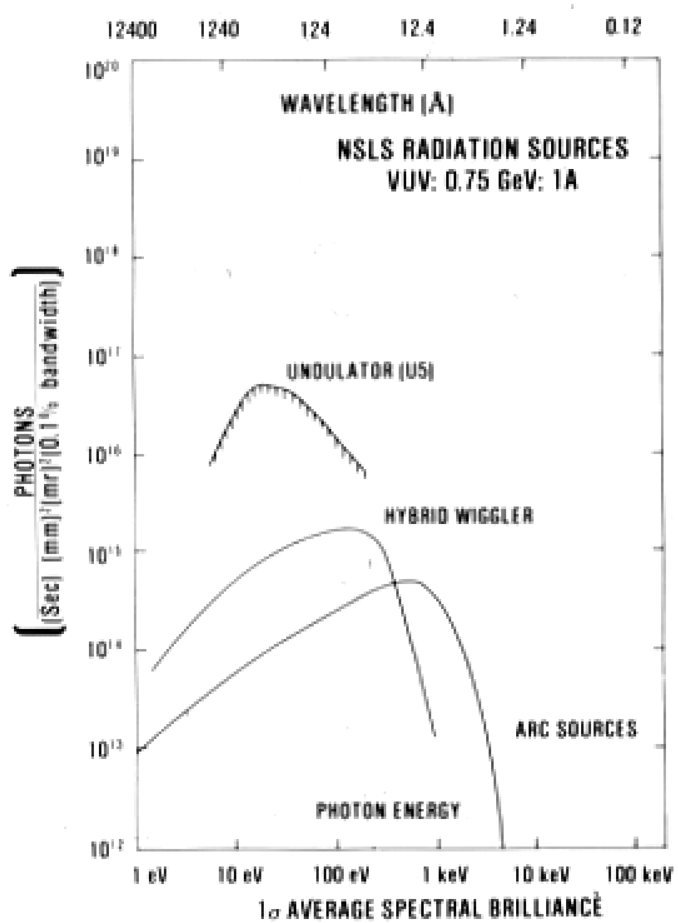
Scattering Power



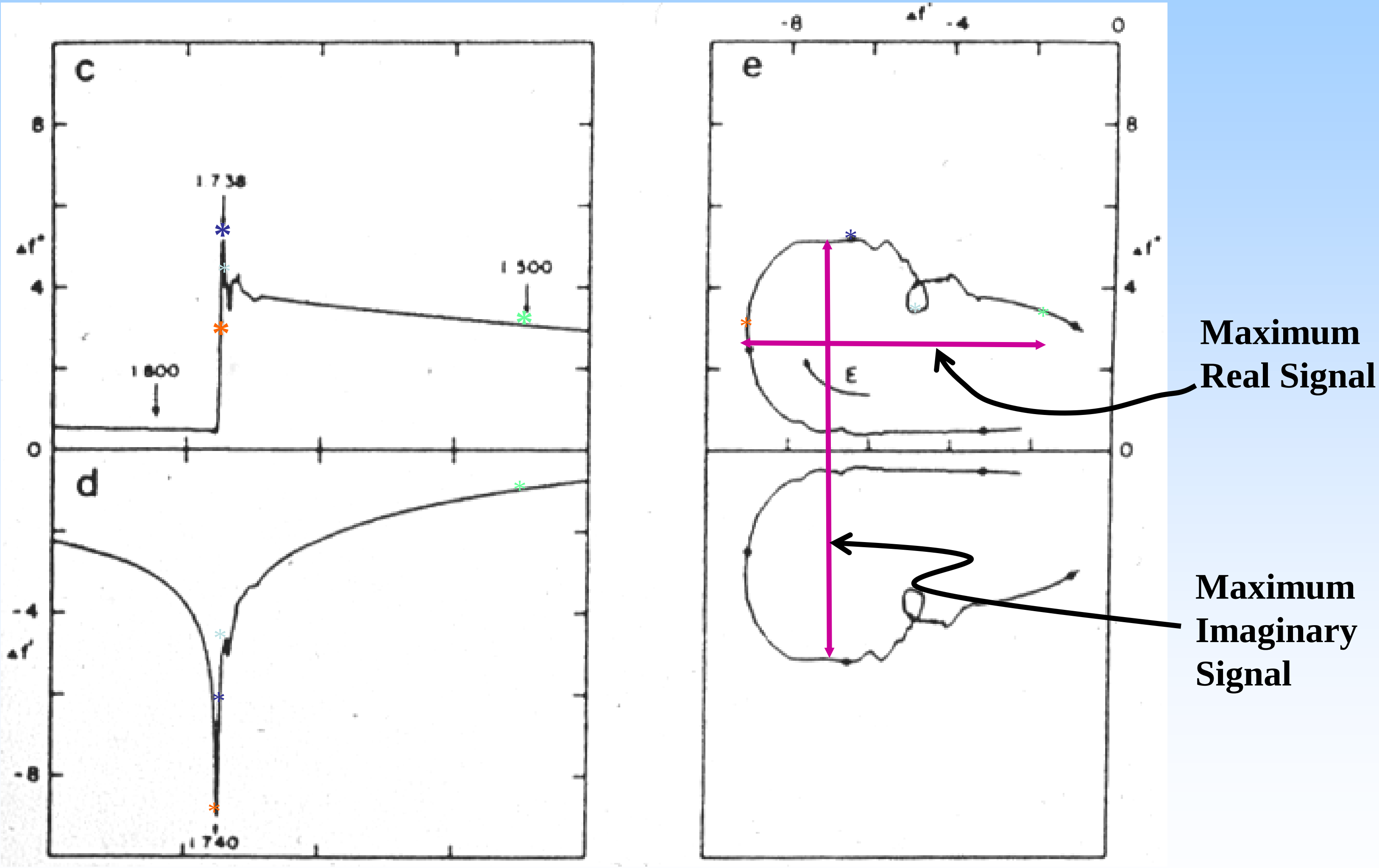
One way to represent this resonance is plots of the shifts in the real part (Δf’) and imaginary part (Δf”) of the scattering of x-rays as a function of the photon energy.

The tunability of the synchrotron source allows us to choose precisely the energy (wavelength) we need.



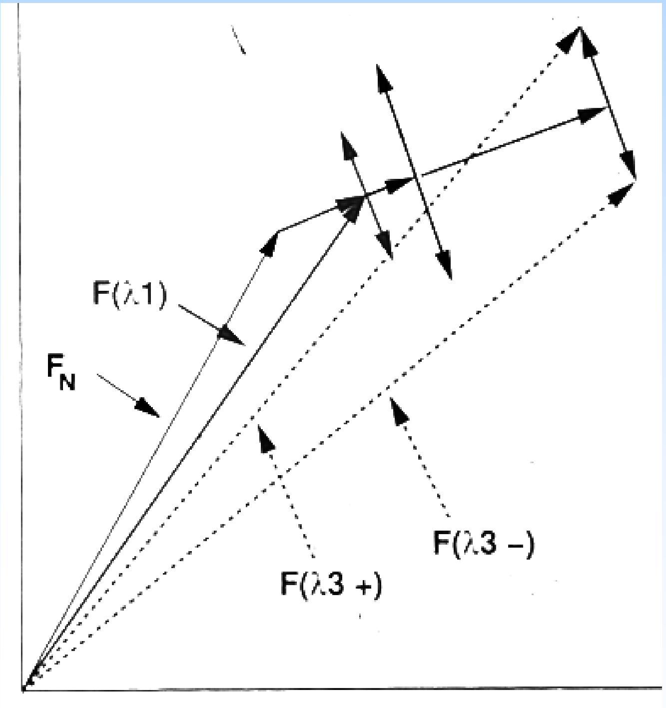


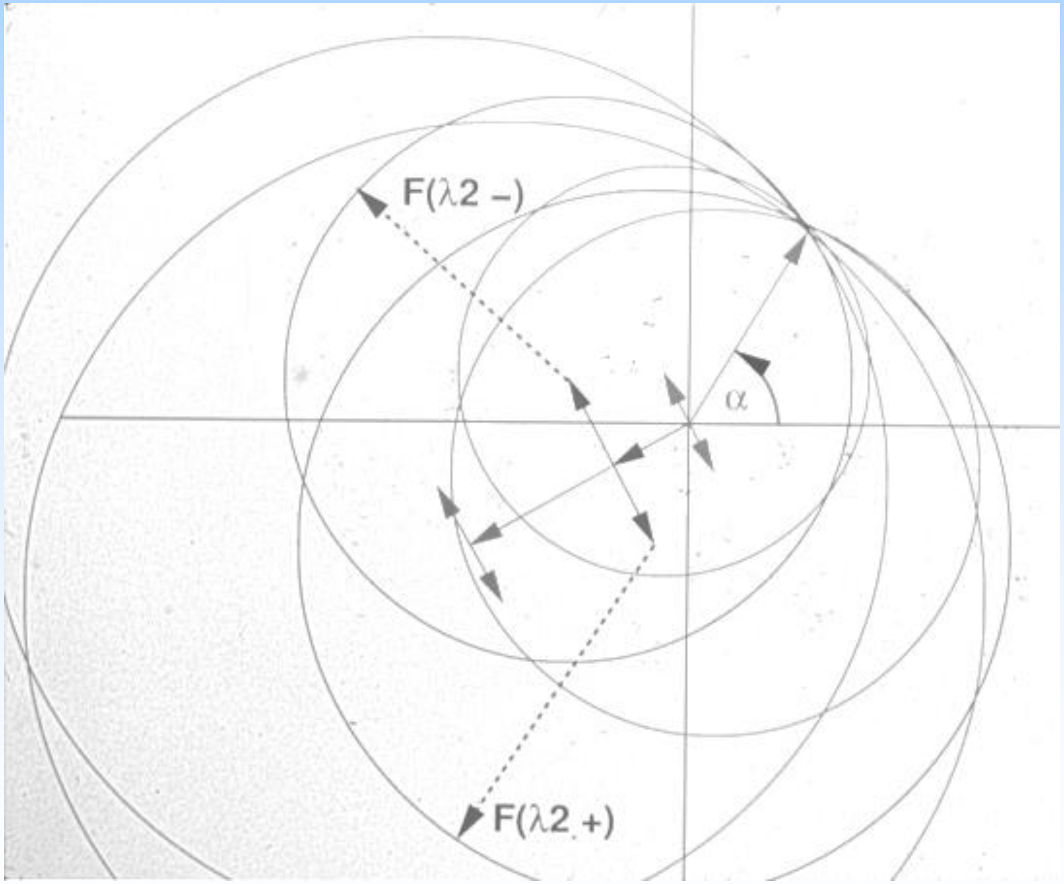
One can see how to choose wavelengths to get large phase contrast for MAD phasing



Spectrum from Phizackerly, Hendrickson, et al. Study of Lamprey Haemoglobin.

This **Multiwavelength Anomalous Diffraction** method often gives very strong phase information and is the source of many new structures.





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How do we find the heavy-atom positions that allow us to do MIR or MAD phasing?

There are generally two methods:

- Patterson-function methods
- Direct-phasing methods

Lindo Patterson saw that to interpret a diffraction pattern, he could correlate the electron density with itself:

We want to show that $P(\vec{u}) = V \int_{\vec{r}} \rho(\vec{r}) \cdot \rho(\vec{r}+\vec{u})\, d\vec{r}$

subs. for $\rho(\vec{r})$

$$P(\vec{u}) = \frac{V}{V^2} \int_{\vec{r}} \left(\sum_{\vec{s}} F(\vec{s}) \exp(-2\pi i\, \vec{r}\cdot\vec{s}) \right) \cdot \left(\sum_{\vec{s}'} F(\vec{s}') \exp(-2\pi i\, \vec{r}\cdot\vec{s}') \right) \exp(-2\pi i\, \vec{s}'\cdot\vec{u})\, d\vec{r}$$

The integral is nonzero only when $\vec{s} = -\vec{s}'$.
So we get

$$P(\vec{u}) = \frac{1}{V} \sum_{\vec{s}} F(\vec{s}) \cdot F^*(\vec{s}) \exp(-2\pi i\, \vec{u}\cdot\vec{s})$$

Also, since $|F(\vec{s})| = |F(-\vec{s})|$

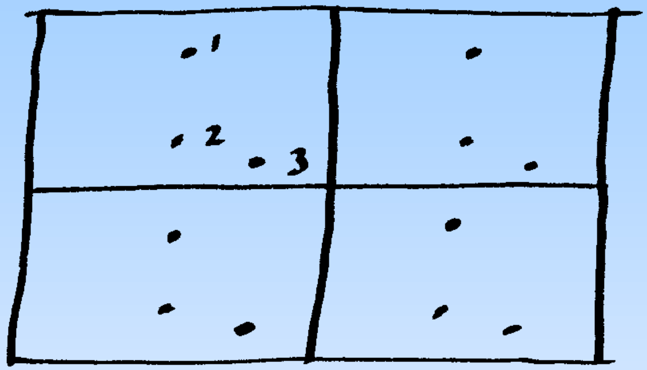
$$P(\vec{u}) = \frac{2}{V} \sum_{\vec{s}=0}^{+\infty} F^2(\vec{s}) \cos(2\pi\, \vec{u}\cdot\vec{s})$$

This is the cosine transform of intensity!

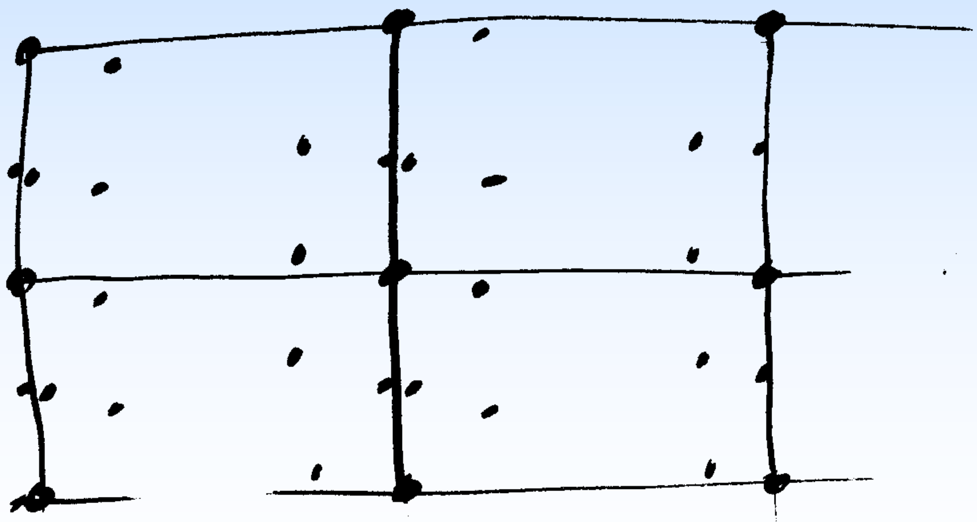
How to interpret the Patterson Function

Consider a structure with 3 atoms

$\vec{r}$-space



$\vec{u}$-space



n^2 peaks total
n at origin
$n(n-1)$ not at origin.

About the same time (all of this happened only a "short" time ago, in the '50s) David Harker saw a neat way to approach "solving" the Patterson function:

The Harker Section

To find out the vectors you will expect — look at equiv. positions:

Example: P2, look at vectors between atoms related by U.C. symmetry

$$(x,y,z) - (\bar{x}, \tfrac{1}{2}+y, \bar{z}) = (2x, \tfrac{1}{2}, 2z)$$

The peak relating x + z will arise at (uvw) = (u, ½, w) — on the v = ½ section.

This method is the basis of software such as HEAVY (Terwilliger)

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To Recapitulate

- **You already knew something** -- lenses, crystals.
- Crystals give ordered arrays of diffraction spots because the **molecules are in ordered arrays**.
- The **Reciprocal Lattice is a mathematical metaphor** for sets of lattice planes that obey Braggs' Law.
- We actually measure diffraction data just by **rotating the crystal** in the x-ray beam and recording diffraction, a lot like a CAT scan.
- Simple mathematics, which turns out to be the **Fourier transform**, allows us to calculate the diffraction pattern
- and, conversely, to calculate the structure from the diffraction.
- The use of heavy atoms, and sometimes resonant effects, allow us to **measure phases** to solve the structures.

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But I can tell you this, if you really want to learn it…

Teach It!